

Figure S1. Maps of plasmids used for *P. tricornutum* transformation. CDS are shown in pink, in green promoters and terminators and fluorescence mVenus gene in yellow.

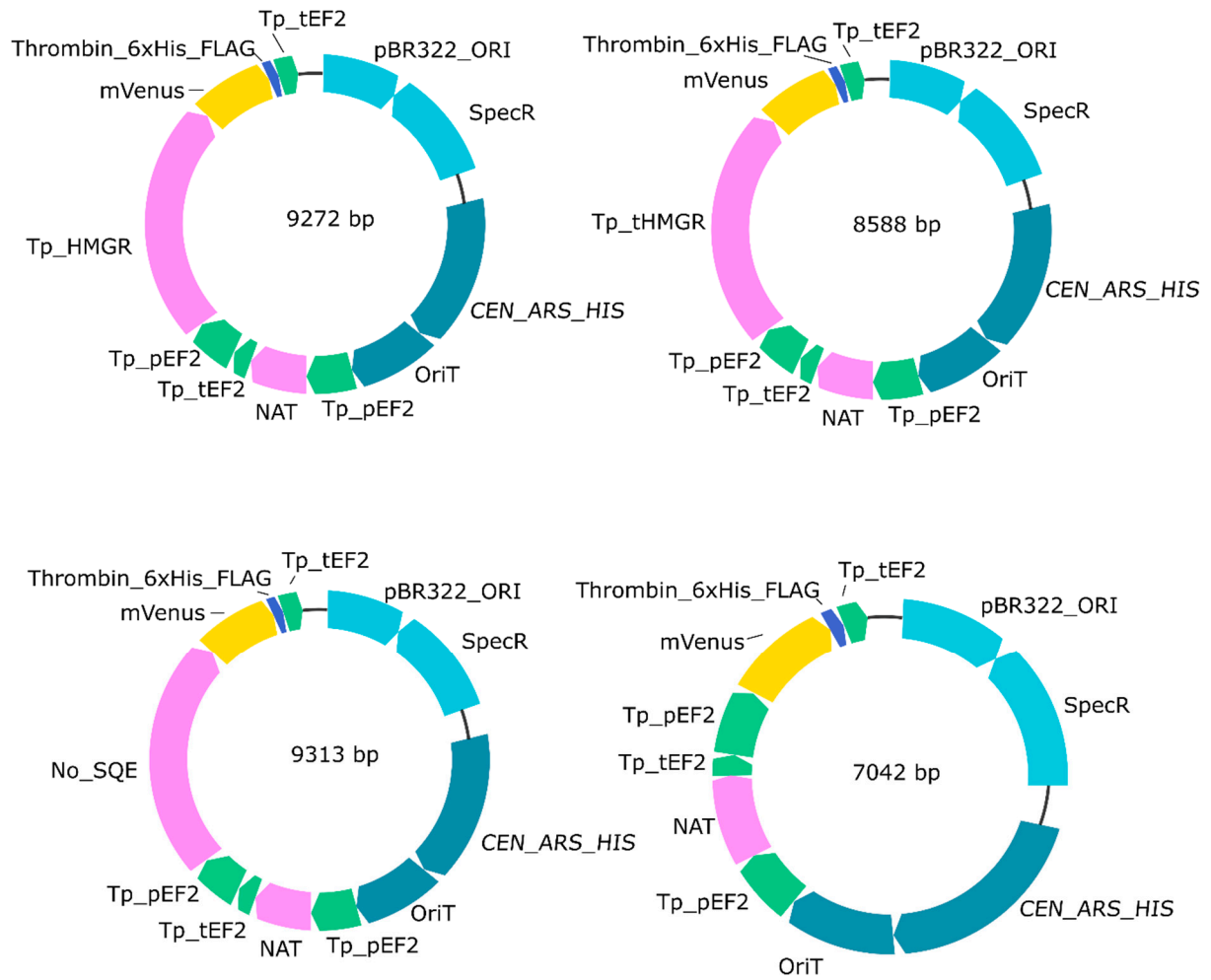


Figure S2. Maps of plasmids used for *T. pseudonana* transformation. CDS are shown in pink, in green promoters and terminators and fluorescence mVenus gene in yellow.

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Thalassiosira_pseudonana 311  VLGANCEIVVGYIPIPVGIVGPVTLNGESVYIIPMATTEG*LVASTNRGCKAITQSSGARSTILRDGITRA
Phaeodactylum_tricornutum 286  VHGANCEIVVGYVPLPVGIVGLVGLTVNGEIVYVPMATTEGCLVASTNRGAKAITAGGGATAVLLRDGITRA
Saccharomyces_cerevisiae 677  VFGACCENVIGYMPLPVGVI GPLVIDGTSYHI PMATTEGCLVASAMRCKAINAGGGATTVLTKDGMTRG
Arabidopsis_thaliana 228  ILGQCCEMPVGYIQIPVGIAGLELLDGYEYSVPMATTEGCLVASTNRGCKAMFISGGATSTVLKDGMTTRA
Homo_sapiens 522  VMGACCENVI GYMPIPVGVAGPLCLDEKEFQVPMATTEGCLVASTNRGCRATIGLGGGASSRVLADGMTRG

Thalassiosira_pseudonana 410  FESTTSFGKLI EASPTVAGRNVIYLRFCFSGDAMGMNISKGSLAVIECLREQF--PQLSLVALSGNMCT
Phaeodactylum_tricornutum 386  FESTTSFGKLLKCAPTVAGRNVIYLRFCFSGDAMGMNIVSKGSLAVIETLQQEF--PELVLVALSGNMCT
Saccharomyces_cerevisiae 777  FNSTSRFARLQHIQTCLAGDLLFMRFRTTGDAMGMNISKGV EYSLKQMV EYGWEDMEVVS VSGNYCT
Arabidopsis_thaliana 328  FNRSSRFARLQSVKCTIAGKNAYVRFCCSTGDAMGMNIVSKGVQNVLE YLTDDF--PDMDVIGISGNFCS
Homo_sapiens 622  FDSTSRFARLQKLH TSIAGRNL YIRFQSRSGDAMGMNISKGTEKALSKLHEYF--PEMQILAVSGNYCT

Thalassiosira_pseudonana 508  TLKTSVPAIVEANVNKNLIGSAMAGTVGGFNAHAANNVTAVFLATGQDPAQNV ESSNCITLMEVSP--EG
Phaeodactylum_tricornutum 484  TLKTTVHSMVQTNLHKNLIGSAMAGALGGFNAHASNIVTAVFLATGQDPAQNV ESSNCITLLEETE--EG
Saccharomyces_cerevisiae 877  VLKSDVSALVELNIAKNLVGSAMAGSVGGFNAHAANLVAVFLALGQDPAQNV ESSNCITLMKEVD---G
Arabidopsis_thaliana 426  VLKTSVAALVELNMLKNLAGSAVAGSLGGFNAHASNIVSAVFIATGQDPAQNV ESSQCITMMEAIN-DGK
Homo_sapiens 720  VLKTTTEAMIEVNIKNLVGSAMAGSIGGYNAHAANIVTAIYIACGQDAAQNV GSSNCITLMEASGPTNE

Thalassiosira_pseudonana 606  AIGVKC-GGENPGDNARQLAHVVCATMAGELSLMAALASNSLVAAHMQHNRKPASK-----
Phaeodactylum_tricornutum 582  AMGVRG-GGATPGAHAQKLAQIVASATLAGELSLAALAANTLVQAHMQHNRKPAK-----
Saccharomyces_cerevisiae 974  LLGVRGPHATAPGTNARQLARIVACAVLAGELSLCAALAAHGLVQSHMTHNRKPAEPTKPNNDATDINR
Arabidopsis_thaliana 525  LLGVKGASTE SPGMNARRLATIVAGAVLAGELSLMSAIAAGQLVRSHMKYNRSSRDISGATTTTTTTT--
Homo_sapiens 820  MLGVQGACKDNPGENARQLARIVCGTVMAGELSLMAALAAHGLVSKHMHNRSKINLQDLQGACTKKTA-

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Figure S3. Sequence alignment of HMGR protein (catalytic domain) from model organisms.

Asterisks (*) indicate conserved catalytic residues.

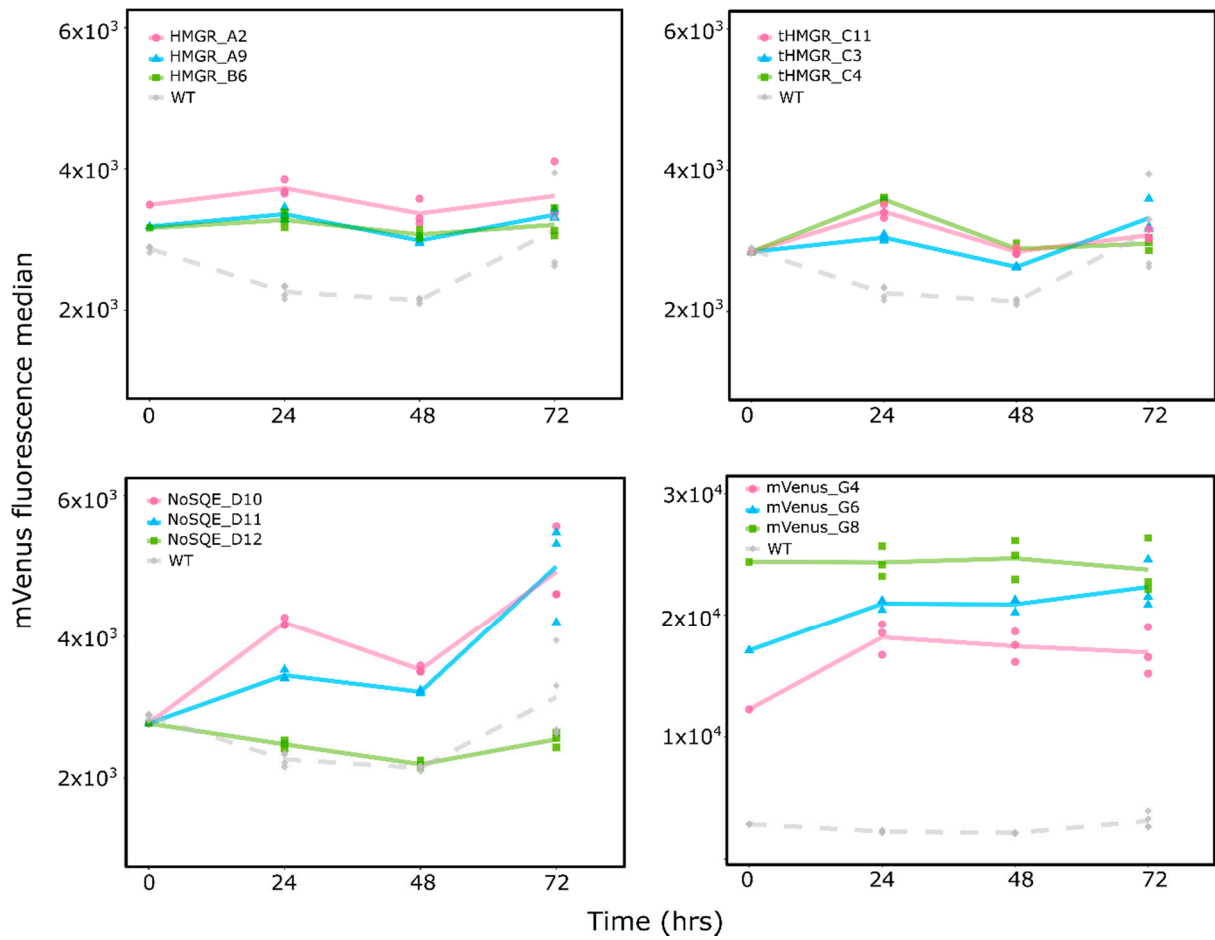


Figure S4. mVenus fluorescence during full scale experiment in *P. tricornutum* transformants (n = 3).

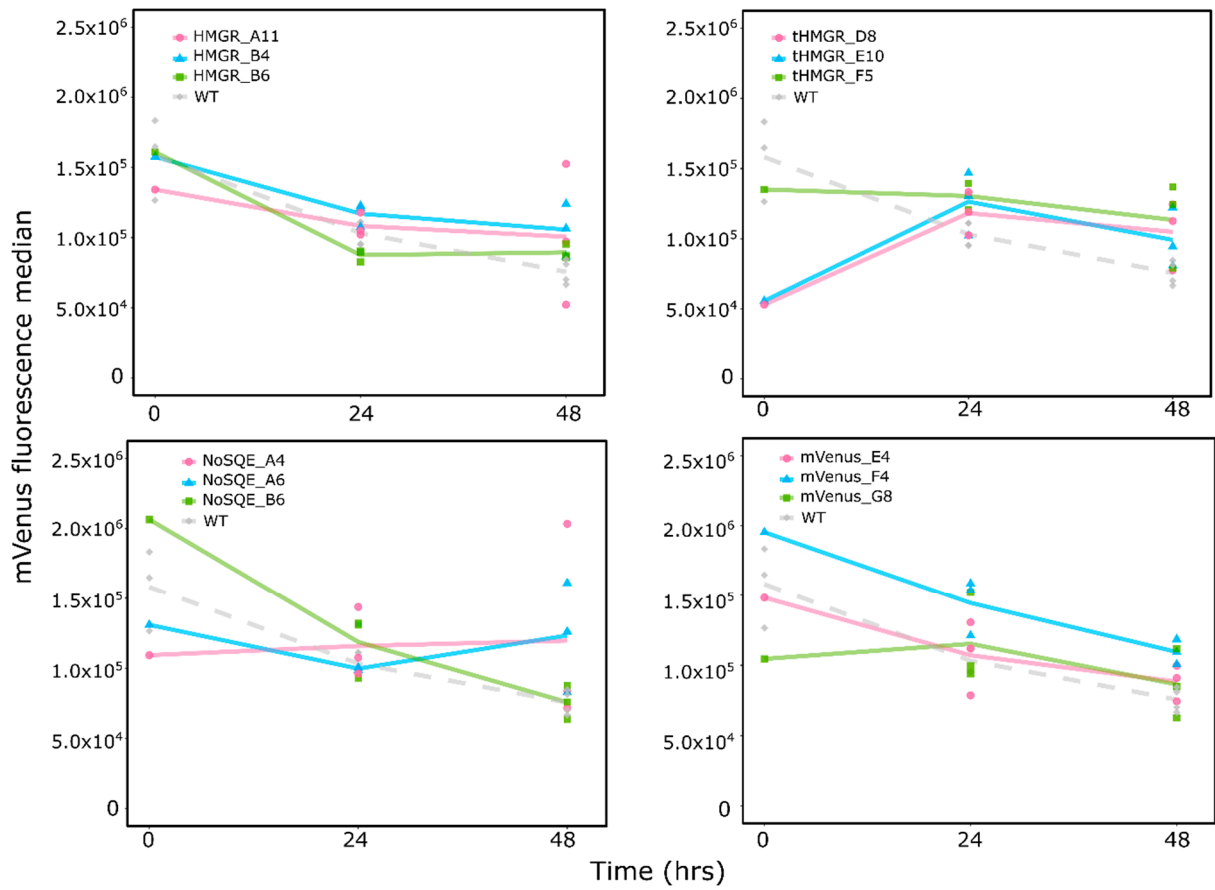


Figure S5. mVenus fluorescence during full scale experiment in *T. pseudonana* transformants (n = 3).

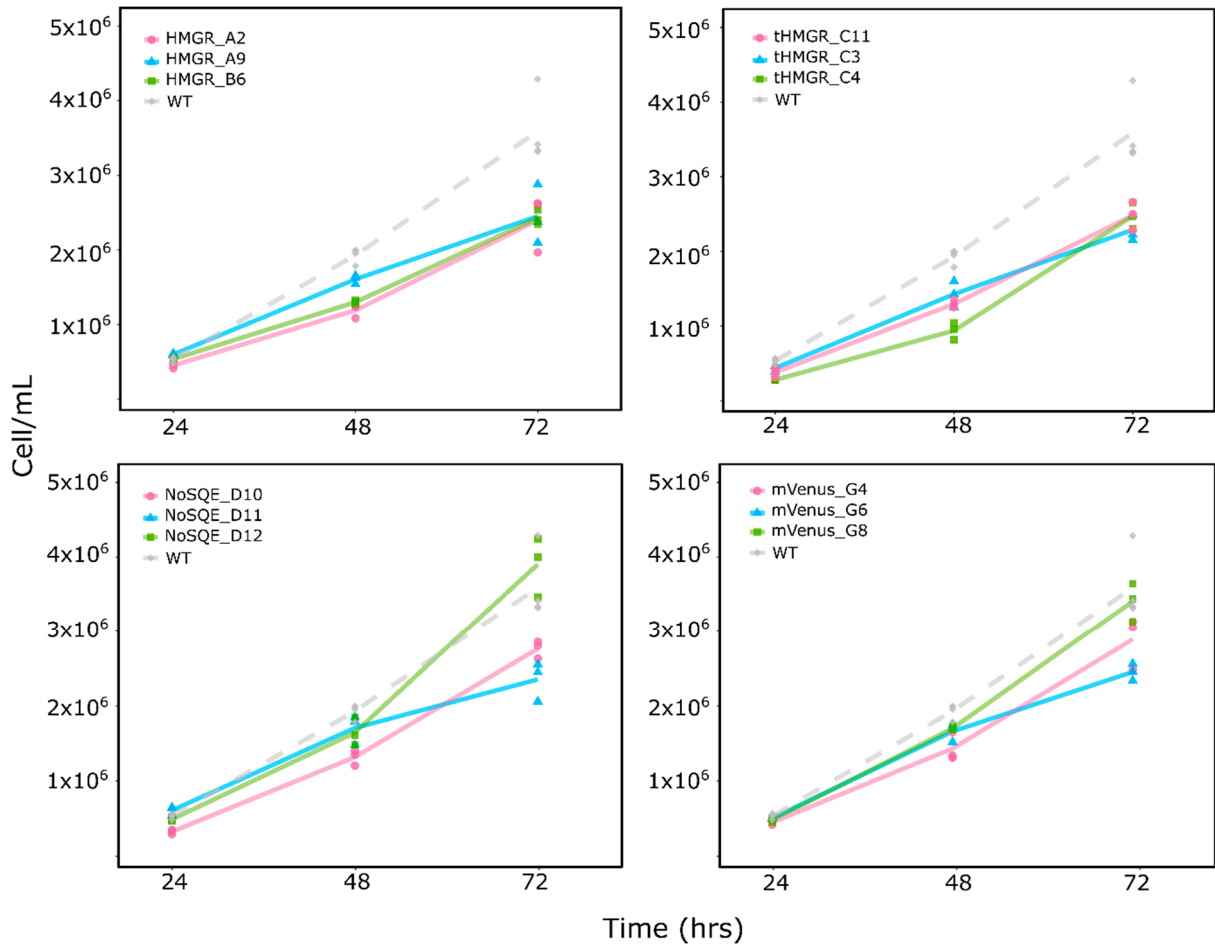


Figure S6. Growth curves during full scale experiment for *P. tricornutum* transformants (n = 3).

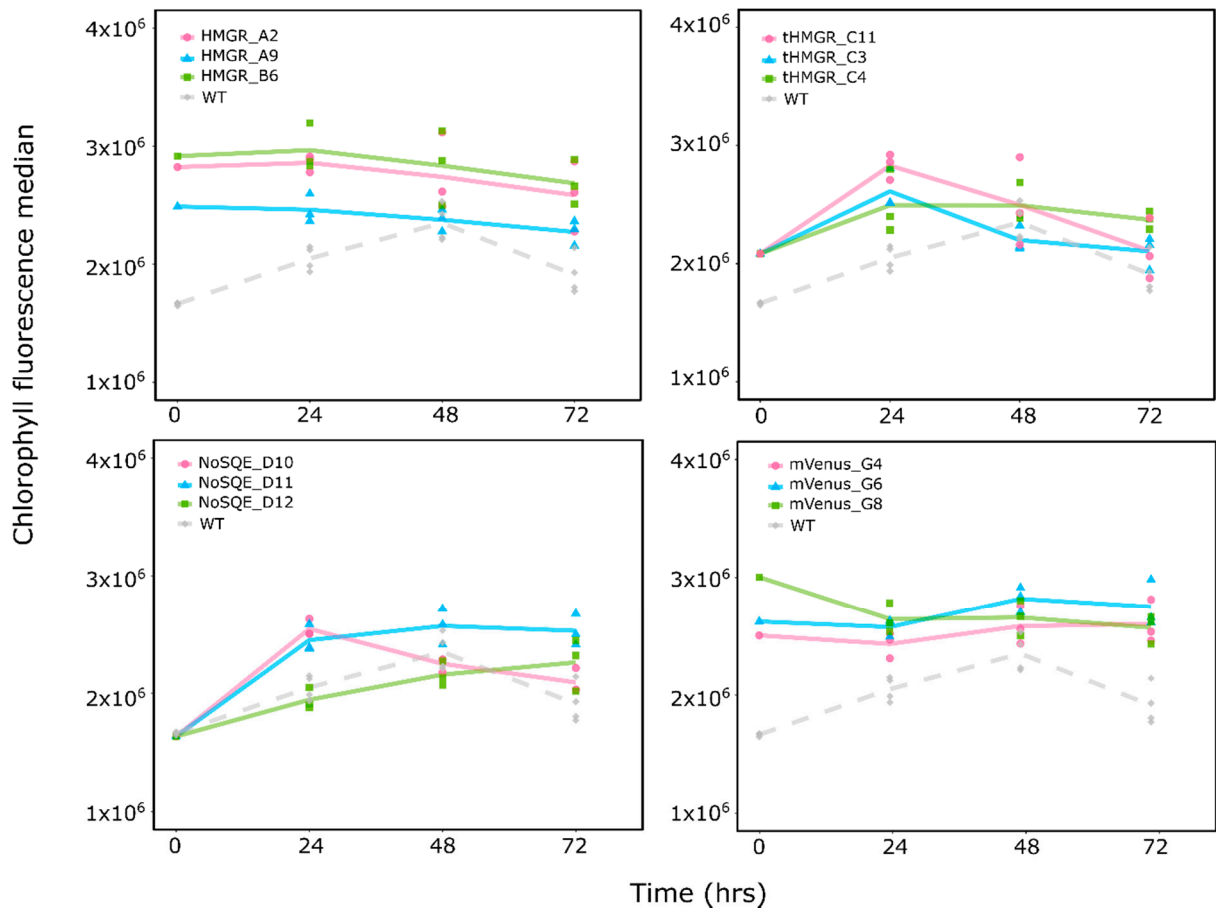


Figure S7. Chlorophyll fluorescence during full scale experiment in *P. tricornutum* transformants (n = 3).

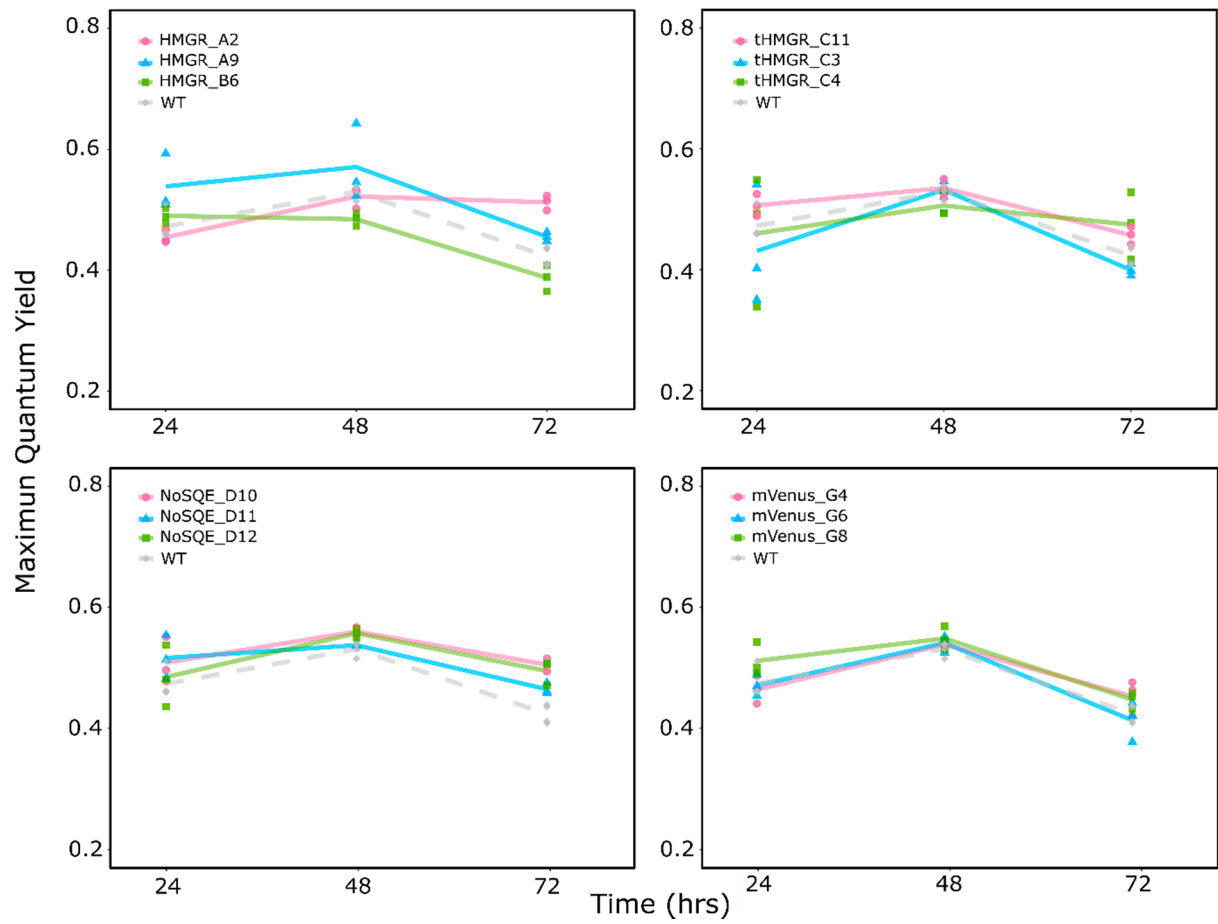


Figure S8. Maximum quantum yield for *P. tricornutum* transformants during full scale experiment (n = 3).

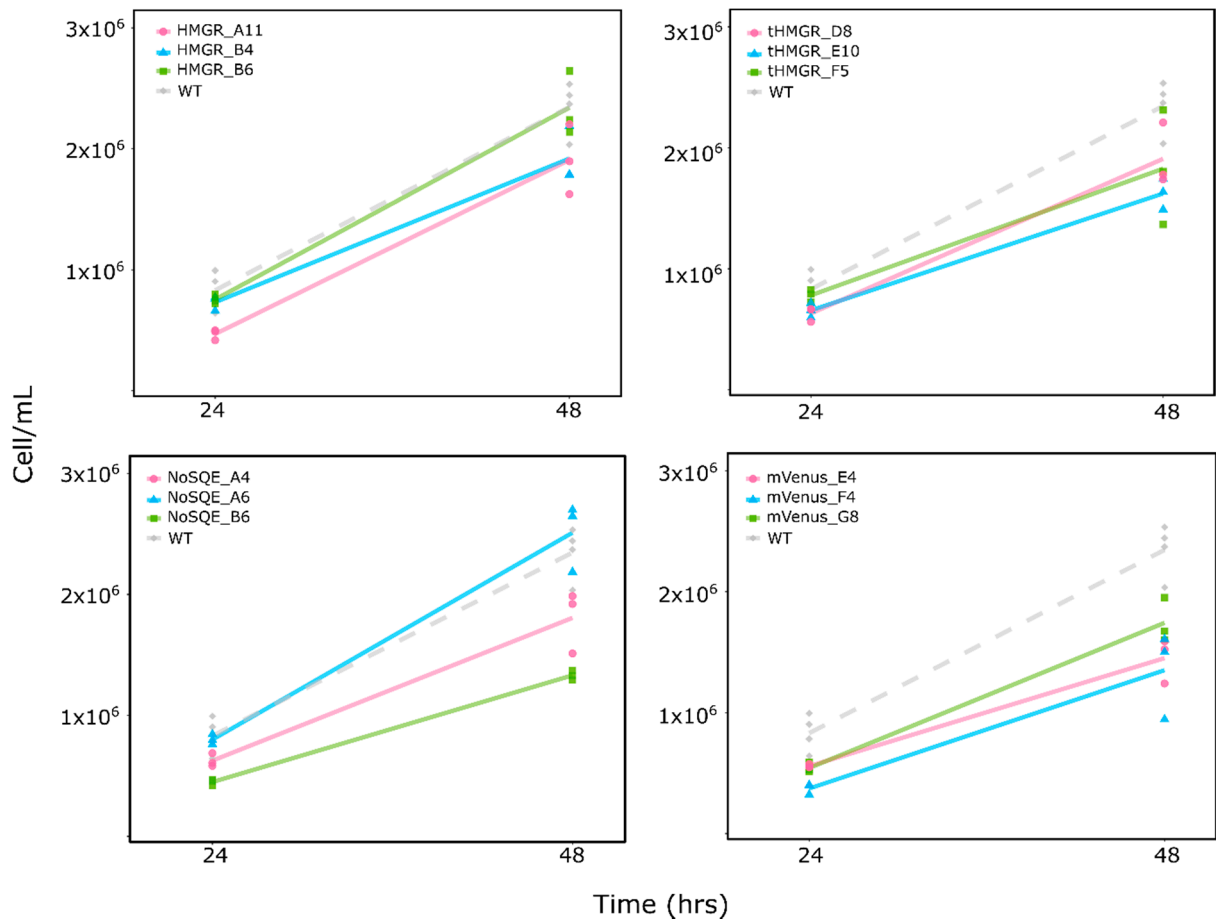


Figure S9. Growth curves during full scale experiment for *T. pseudonana* transformants (n = 3).

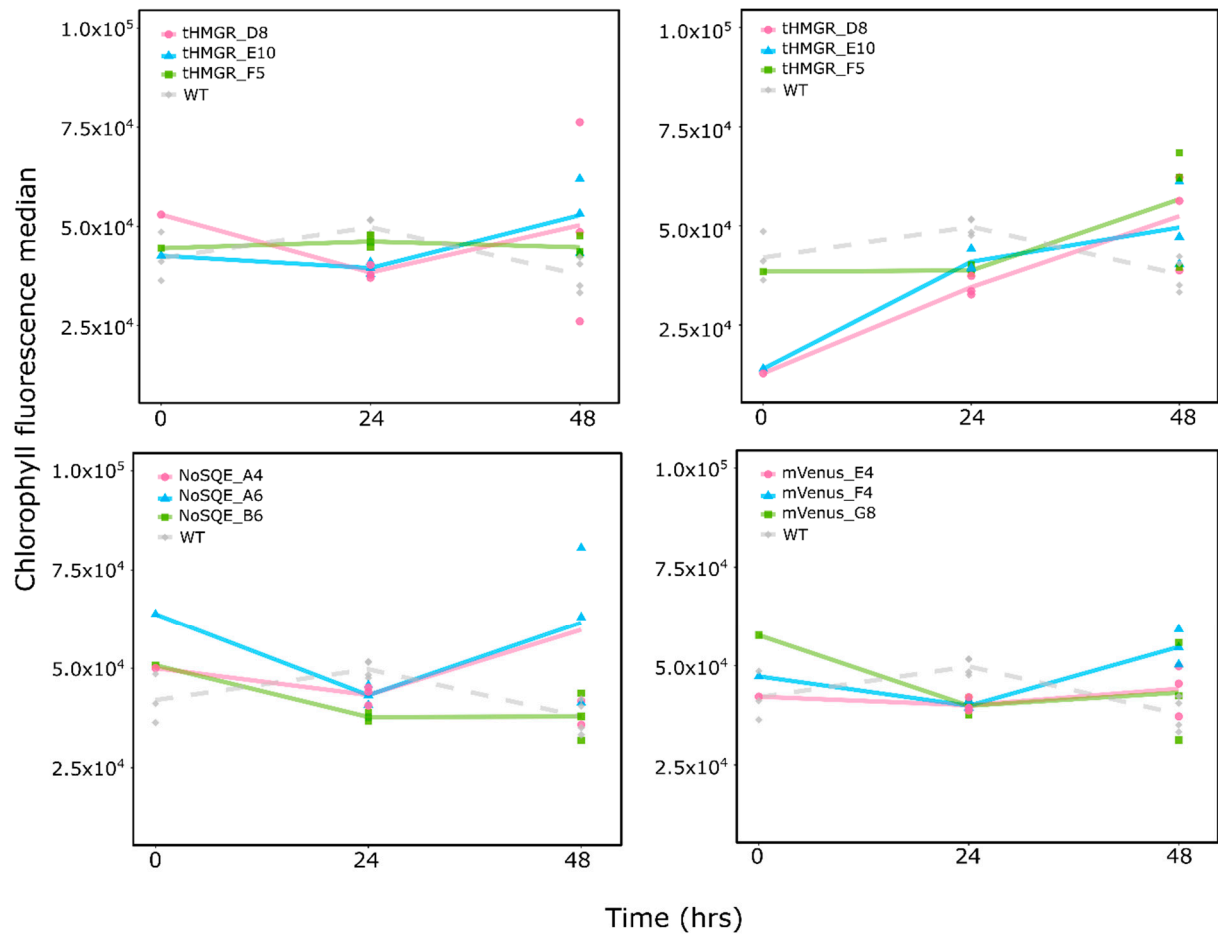


Figure S10. Chlorophyll fluorescence during full scale experiment in *T. pseudonana* transformants (n = 3).

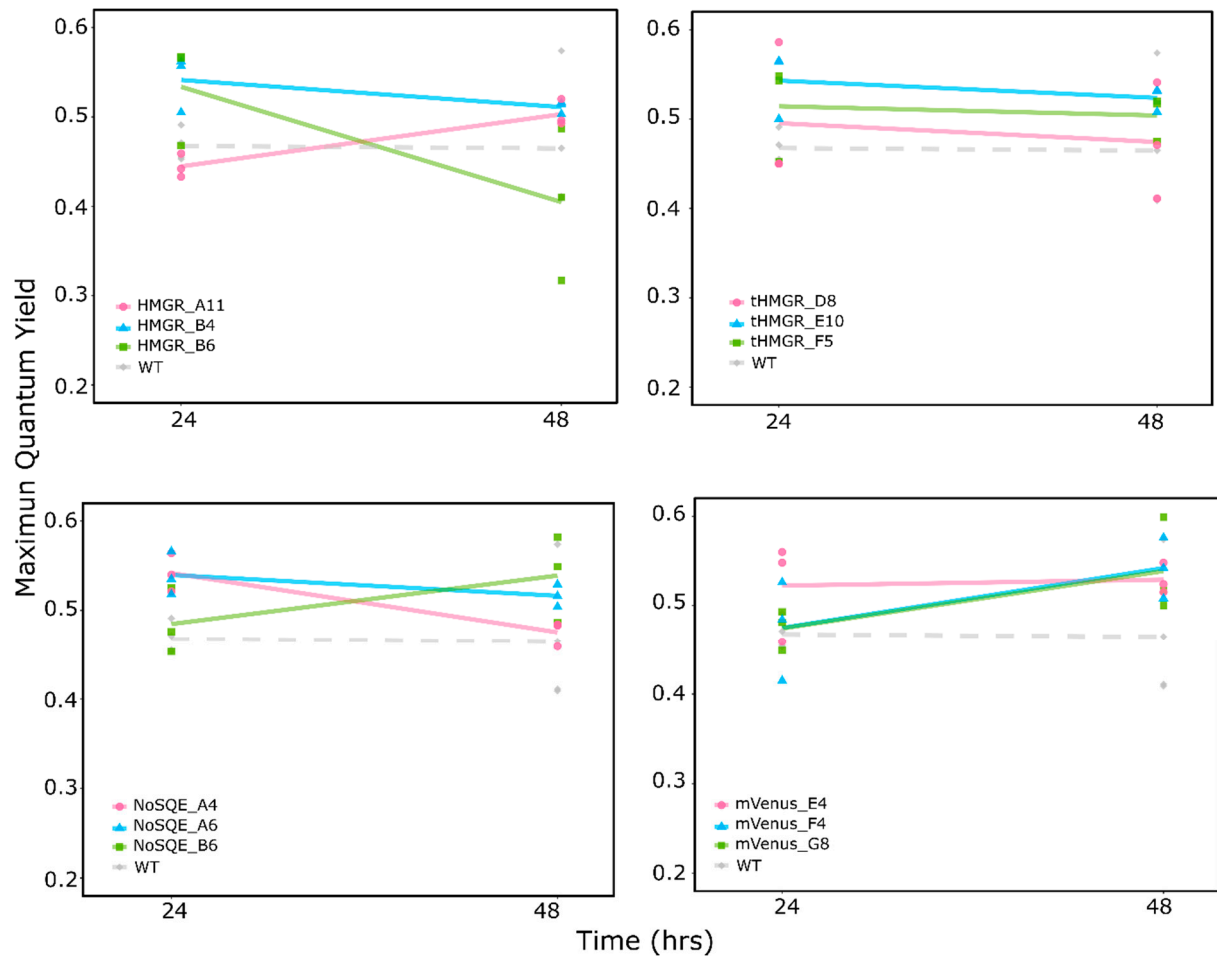


Figure S11. Maximum quantum yield for *T. pseudonana* transformants during full scale experiment (n = 3).

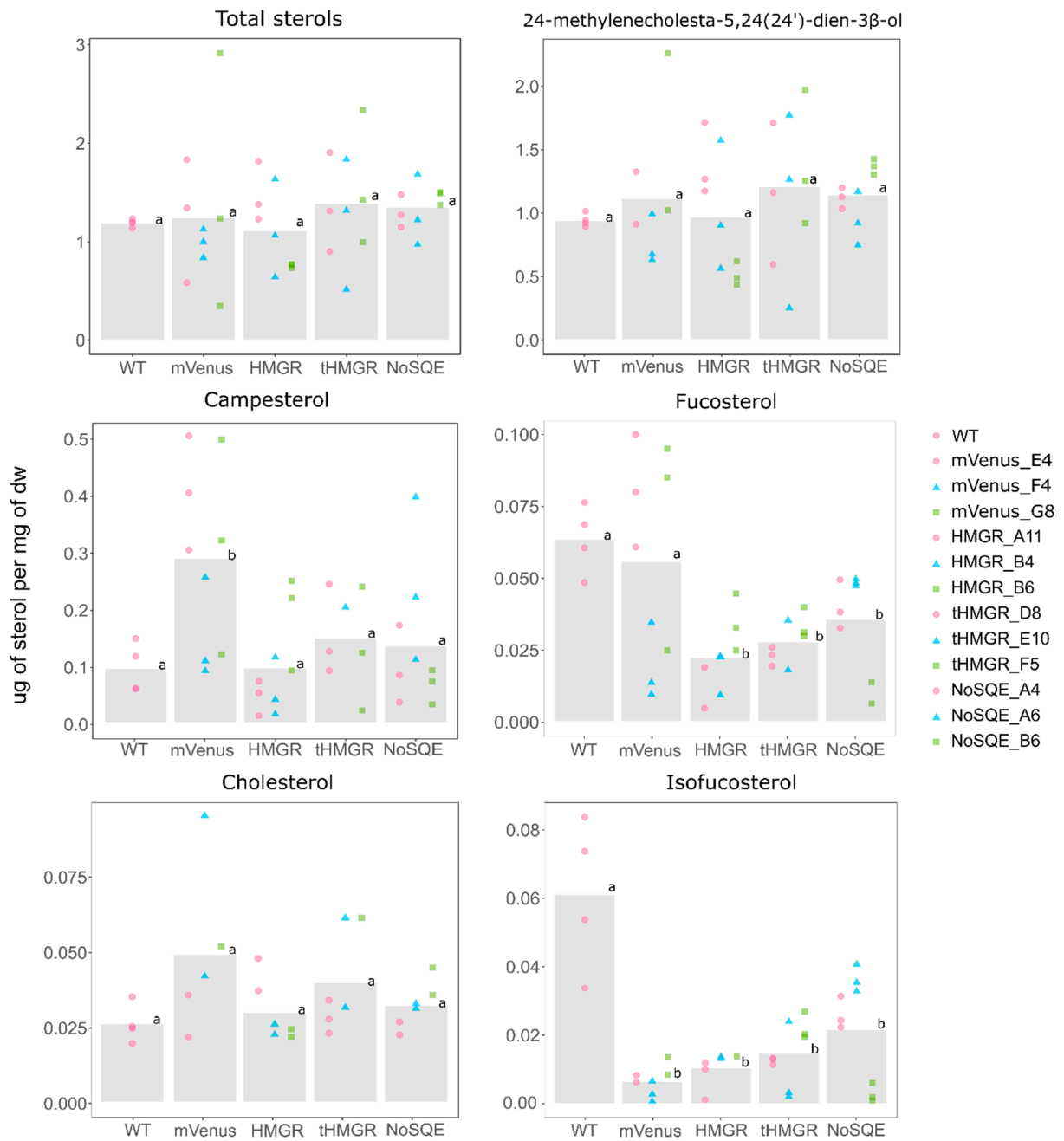


Figure S12. Sterol levels in *T. pseudonana* transformants. Identical letters denote no statistically significant differences among groups using the Pairwise Wilcoxon Rank Sum tests ($p < 0.05$, $n = 9$).

Table S1 L0 parts for construction of episomes using uLoop assembly method [62]. Primers sequences used for domestication are presented for L0 each part

L0 part	Gene ID	Description	Template	Primers	Plasmid number
AC_pTpEF2	269148	Elongation factor promoter	<i>P. tricornutum</i> genomic DNA	F: AAGCTCTTCATCCggagAGTGTGCAATGCAGTCAATTCAATAGATATG R: TTGCTCTTCTTCGcattCTTGACGTTCTTTTCTCTTTAATTAATCGCG	
AC_pPtEF2	Phatr3_J35766	Elongation factor promoter	<i>P. tricornutum</i> genomic DNA	F: AAGCTCTTCATCCggagTCCATTTTGACATGTTTCCTAGCTAGAAG R: TTGCTCTTCTTCGcattTGTGTGGAGAGAACGAGCAGCAGCG	
AC_pPt49202	Phatr3_J49202	Predicted protein JCVI	NA	NA	
AC_CENARSHIS	NA	CEN6-ARSH4-HIS3 episome maintenance sequence from PtPBR11 (Genebank KX523203) (Diner <i>et al.</i> , 2016)	NA	NA	
CF_OriT	NA	Origin of transfer, originally called basis of mobilization (Diner <i>et al.</i> , 2016)	PtPBR11 (Genebank KX523203)	F: CAGAAGCTCTTCATCCaatgGATCGTCTTGCCTTGCTCGTCGG R: TTGCTCTTCTTCGagcATCTTCCGCTGCATAACCCTGCTTCGG	139948
CD_NoSQE	521007	0.0	NA	NA	
CD_TpHMGR	33680	3-hydroxy-3-methylglutaryl-coenzyme A reductase (EC: 1.1.1.34)	<i>T. pseudonana</i> genomic DNA	F: AAGCTCTTCATCCaatgGCGGCAGCAGCAGCACCACCATAG R: TTGCTCTTCTTCGacctgaCTTTGAAGCAGGCTTGCGATTATGTTG	
CD_TptHMGR	33680	Truncated 3-hydroxy-3-methylglutaryl-coenzyme A reductase (EC: 1.1.1.34)	<i>T. pseudonana</i> genomic DNA	F: AAGCTCTTCATCCaatgAGCCCCAACGACCCACCCGTCAAAG R: TTGCTCTTCTTCGacctgaCTTTGAAGCAGGCTTGCGATTATGTTG	
CD_PtHMGR	Phatr3_J16870	3-hydroxy-3-methylglutaryl-coenzyme A reductase	<i>P. tricornutum</i> genomic DNA	F: AAGCTCTTCATCCaatgACGGTGACTATCAGCAGTAGTATTAG R: TTGCTCTTCTTCGacctgaCTTGCGGCGGGTTTGCGGTTG	
CD_PttHMGR	Phatr3_J16870	Truncated 3-hydroxy-3-methylglutaryl-coenzyme A reductase	<i>P. tricornutum</i> genomic DNA	F: AAGCTCTTCATCCaatgGACTCTATTTCCACCAAGACCAGCGCG R: TTGCTCTTCTTCGacctgaCTTGCGGCGGGTTTGCGGTTG	

CD_bsd	NA	blasticidin-S deaminase	pST1374-N-NLS-flag-linker-Cas9-D10A (Addgene #51130)	F: AAGCTCTTCATCCaatgGCCAAGCCTTTGTCTCAAGAAGAATCCAC R:TTGCTCTTCTTCGacctgaGCCCTCCCACACATAACCAGAGGG	
CD_nat	NA	nourseothricin acetyltransferase	pAGM4723:TpCC_Urease (Addgene #85982)	F: AAGCTCTTCATCCaatgACCACTCTTGACGACACGGCTTACCGG R:TTGCTCTTCTTCGacctgaGGGGCAGGGCATGCTCATGTAGAGCG	
CD_mVenus		Yellow fluorescent protein	NA	NA	
DE_Thr6XHisFLAG	NA	General C-terminal protein tag. Thormbine, 6xHIS and FLAG (DYKDDDDK) tag	DE_Venus-ThrHISFLAG LO part	F: AAGCTCTTCATCCaggtGCGCTGGTCCCTCGCGGTAG R: TTGCTCTTCTTCGagcTCACTTATCGTCATCATCCTTGTAGTCG	
DE_Venus-ThrHISFLAG	NA	Yellow fluorescent protein with a Thormbine, 6xHIS and FLAG (DYKDDDDK) tag JCVI	NA	NA	
DE-3xStop	NA	Three stop codon	NA	NA	
EF_tPtEF2	Phatr3_J35766	Elongation factor terminator	<i>P. tricornutum</i> genomic DNA	F: AAGCTCTTCATCCGCTTACAGAAAAACAGACTCATAGGGTAC R:TTGCTCTTCTTCGAGCGGAGACATTACTCCACACGATGG	
EF_tTpEF2	269148	Elongation factor terminator	<i>T. pseudonana</i> genomic DNA	F: AAGCTCTTCATCCgcttATATCTTCTTGCAACAATGGTAGCG R: TTGCTCTTCTTCGagcgAGCAGGGTTGGTTAGAGATAACTAATG	
EF_tPt49202	Phatr3_J49202	Predicted protein	NA	NA	

Table S2. Source and number of transmembrane domains of *HMGR* sequences used for phylogenetic and domain analysis

Organism	MMETSP ID	Present	Source	Strain	Gene ID	TM*
<i>Attheya septentrionalis</i>	MMETSP1449	Y	Transcriptomics	CCMP2084	Transcript_1390	3
<i>Chaetoceros affinis</i>	MMETSP0091	Y	Transcriptomics	CCMP159	Transcript_17836	0
<i>Chaetoceros brevis</i>	MMETSP1435	N	Transcriptomics	CCMP164	NA	NA
<i>Chaetoceros curvisetus</i>	MMETSP0716	N	Transcriptomics	Unknown	NA	NA
<i>Chaetoceros debilis</i>	MMETSP0149	N	Transcriptomics	MM31A-1	NA	NA
<i>Chaetoceros dichaeta</i>	MMETSP1447	Y	Transcriptomics	CCMP1751	Transcript_4115	0
<i>Chaetoceros muelleri</i>	NA	N	Transcriptomics	CCMP1316	NA	NA
<i>Chaetoceros neogracile</i>	MMETSP0752	Y	Transcriptomics	CCMP1317	Transcript_689	3
<i>Chaetoceros sp.</i>	MMETSP1429	Y	Transcriptomics	UNC1202	Transcript_18209	0
<i>Coscinodiscus wailesii</i>	MMETSP1066	Y	Transcriptomics	CCMP2513	Transcript_2372	3
<i>Cylindrotheca closterium</i>	MMETSP0017	Y	Transcriptomics	KMMCC:B-181	Transcript_33356	3
<i>Ditylum brightwelli</i>	MMETSP1001	Y	Transcriptomics	GSO105	Transcript_14074	3
<i>Extubocellulus spinifer</i>	MMETSP0696	Y	Transcriptomics	CCMP396	Transcript_715	3
<i>Fistulifera solaris</i>	NA	Y	Genome	JPCC DA0580	GAX29119.1	3
<i>Fragilariopsis kerguelensis</i>	MMETSP0909	Y	Transcriptomics	L2-C3	CAMPEP_0196091342	2
<i>Homo sapiens</i>	NA	Y	Genome	NA	3156	5
<i>Leptocylindrus danicus</i>	MMETSP0321	Y	Transcriptomics	B650	Transcript_1399	2
<i>Nitzschia punctate</i>	MMETSP0747	Y	Transcriptomics	CCMP561	Transcript_27671	3
<i>Nitzschia sp</i>	MMETSP0014	Y	Transcriptomics	RCC80	Transcript_11673	3
<i>Odontella Sinensis</i>	MMETSP0160	Y	Transcriptomics	Grunow 1884	Transcript_15934	3
<i>Oryza sativa</i>	NA	Y	Genome	NA	LOC_Os02g48330	0
<i>Phaeodactylum tricornutum</i>	NA	Y	Genome	CCMP632	Phatr3_J16870	3
<i>Pseudonitzschia arenysensis</i>	MMETSP0329	Y	Transcriptomics	B593	CAMPEP_0116128146	3
<i>Pseudonitzschia delicatissima</i>	MMETSP0327	Y	Transcriptomics	B596	CAMPEP_0116102328	3
<i>Pseudonitzschia multiseriis</i>	NA	Y	Genome	CLN-47	288249	3
<i>Pseudonitzschia pungens</i>	MMETSP1061	Y	Transcriptomics	cf. pungens	Transcript_13267	3
<i>Rhizosolenia setigera</i>	MMETSP0789	Y	Transcriptomics	CCMP1694	CAMPEP_0178949462	3
<i>Saccharomyces cerevisiae</i>	NA	Y	Genome	ATCC 204508	854900	7
<i>Skeletonema marinoi</i>	MMETSP1428	Y	Transcriptomics	UNC1201	Transcript_20411	3
<i>Skeletonema menzelii</i>	MMETSP0603	Y	Transcriptomics	CCMP793	Transcript_7316	3
<i>Solanum lycopersicum</i>	NA	Y	Genome	NA	Solyc02g082260.3	2
<i>Thalassiosira miniscula</i>	MMETSP0737	Y	Transcriptomics	CCMP1093	Transcript_23358	2
<i>Thalassiosira oceanica</i>	NA	Y	Genome	CCMP1005	91521	2
<i>Thalassiosira pseudonana</i>	NA	Y	Genome	CCMP1335	269148	3

<i>Thalassiosira punctigera</i>	MMETSP1067	Y	Transcriptomics	Tpunct2005C2	Transcript_37597	3
<i>Thalassiosira rotula</i>	MMETSP0403	Y	Transcriptomics	CCMP3096	Transcript_15672	2
<i>Thalassiosira weissflogii</i>	MMETSP1414	Y	Transcriptomics	CCMP1010	Transcript_6235	3
<i>Thalassiothrix antarctica</i>	MMETSP0152	Y	Transcriptomics	L6-D1	Transcript_7954	3

*TM indicates number of transmembrane domains predicted by TMHMM Server v. 2.0 (<http://www.cbs.dtu.dk/services/TMHMM-2.0/>). One of the three transmembrane domains in *Arabidopsis* is located in C-terminus domain.

HMGR sequences for phylogenetic reconstruction

>Thalassiosira_pseudonana

MAAAAAPTIGQRLDTLLAALSNI GDMNHLLSNYQPSTSQIYSLVIVLSVAFSFRLLNSGDDLGSKLSSTVEQ
AKRGEKGTSSKYAKANNKNNKQSYDFPQPKWHILKLTNYVVTTFLLSIITFLSNASVYLNDNTALMTFLGV
WSGLLCYFFGFFGISFVELDDLVTADAGQQRQQGVVTQPQKQASCKSRKVESPNDPPVKVISLHPPASS
TPVCS DPIKSKSPTNTGIPTNVKDL SNEEIATLVLQDKIKDHQLEKLLDPHRAVAVRRLKFDALLSSLGKTTGE
DDKGGV LSELPEHDL DYKRVLGANCEIVVGYIPIVGVIPVTLNGESVYIPMATTEGLVASTNRGCKAI
TQSGGARSTILRDGITRAPCVRLPSAHEAAQVHLWIEEADNFAKLKEAFESTTSFGKLIASPTVAGRNVIYR
LRCFSGDAMGMNMISKGSLAVIECLREQFPQLSLVALSGNMCTDKKAAAMN WIEGRGKSVVIEATIPKD
VVRSTLKTSPVAIVEANVNKNLIGSAMAGTVGGFNHAHAANNVTAVFLATGQDPAQNVESNCITLMEVS
PEGDLWISCTMPSIEVGT VGGGTGLSAQSACLAIGVKGGGENPGDNARQLAHVVACATMAGELSLMA
ALASNSLVAAHMQHNRKPASK

>Phaeodactylum_tricornutum

MTVTISSISSISSIGSSTTAPT LGMQVDALVQQMDNLSSTQLYGLIVGLTILVSFVLLGSSADIPVSLRDT
TKETSSSSSTPRKTTT VSSSSNRGPEPRWHIFTYVNYAIVACFVASVAEFGRNASAYLAADDNVVLYFLV
AWSVFLCYFFGFFGVSVFHDADA AVASPTPTKPTVRDSISTKTSASNTLSARHPPAPSAPVCS DPSSFTP
IHTSKTNITLDNAAICQLVLTNQIKDHELEKRLDAHRAVQVRRLLVVAHKLDTLEHINAHALDNL PSEPS
LDYTRVHGANCEIVVGYVPLPVGLVGLTVNGETVYVPMATTEGCLVASTNRGAKAITAGGGATAVLLRD
GITRAPCVRMP SAAQAHLKLCWCETPQH FSTLKRAFESTTSFGKLLKCAPTVAGRNVIYRLTCFSGDAMG
MNMVSKGSLAVIETLQQEFPELV LVALSGNMCTDKKAAATNWLEGRGKSIVVEATIPKDVVTNTLKT TVH
SMVQTNLHKNLIGSAMAGALGGFNHASNIVTAVFLATGQDPAQNVESNCITLLEETEEDLWISCTM
PSIEVGT VGGGTSLPAQAACLQAMGVRGGGATPGAHAQKLAQIVASATLAGELSLLAALAANTLVQA HM
QHNRKPAAK*

>Fistulifera_solaris

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>Pseudonitzschia_multiseries

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AVFLATGQDPAQNVESNCITLMEEQDDGDLWMCCTMPSIEVGT VGGGTSLPAQAACLEAIGCKGGGS
TPGANAKLATVVAATMAGELSLLAALAANTLVQA HMVHNRKPAAK*

>Thalassiosira_oceanica

MMAGTATIGQRDLALVATAQANTDGSAAALFALVIGISVSFSFYLLNSGSGSTASAEMTKCGAQLSTCPKD
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>Extubocellulus_spinifer

MTTVPTLGERLDALVDRIDSIPTQLYIAAVVTSVVFSCMLLNSGGSNGVDPKIPDNI
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SLAVIDLLKSTFPTLELVALSGNVCCDKKSGAINWIEGRGKSVVVEATIPLEVVRSTLKT
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>Attheya_septentrionalis

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>Thalassiosira_weissflogii

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>Odontella_sinensis

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R F A S D A S R H M S D S T S L L K F M S I W S A L L C Y F F G F G I S F V D A E G F S V A G E E T K T T E K T T K K
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E P S L D Y G R V H G A N C E I V V G Y V P L P C G V I G P L T L D G E T V Y V P M A T T E G C L V A S A N R G C K A I
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G K

>Rhizosolenia_setigera

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S P V A A V H K K Q V S K A A S F S R P I H P P A M S T H P V C G T G E I S S P T T T A S L P D D I Q S L S N E V A A S
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C E I V V G Y V P I P V G M C G P I T L N G E S V Y I P M A T T E G C L V A S T N R G C K A I S A G S G A T S V I L R D
G I T R A P C L R M K S A K E A A D L K L W C E E Q E N F L L L K X A F E S T T S F G K L L S A E P T V S G K N V Y X R
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D P A Q N V E S S N C I T L L E E T E D G D L W I C C T M P S I E V G T V G G G T G L P A Q S S C L K M I G C K G G G E
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>Coscinodiscus_wailesii

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>Skeletonema_marinoi

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>Skeletonema_menzelii

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>Thalassiosira_miniscula

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>Thalassiosira_punctigera

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>Ditylum_brightwellii

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>Cylindrotheca_closterium

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>Nitzschia_punctata

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>Nitzschia_sp.

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>Pseudonitzschia_pungens

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>Saccharomyces_cerevisiae

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>Arabidopsis_thaliana

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>Homo_sapiens

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>Oryza_sativa

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>Solanum_lycopersicum

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>Thalassiosira_rotula

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>Pseudonitzschia_delicatissima

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>Pseudonitzschia_arenysensis

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>Fragilariopsis_kerguelensis

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>Leptocylindrus_danicus

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>Chaetoceros neogracile

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>Chaetoceros dicheta

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>Chaetoceros affinis

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