

Supplement

Growth and nutrient uptake characteristics of *Heterosigma akashiwo* (Raphidophyceae) under nitrogen and phosphorus concentrations in the East China Sea

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Table S1. Artificial seawater recipe [1].

Component	stock solution [g L ⁻¹]	Final concentration in media	supplier
Salt solution I – anhydrous salts			
NaCl	21.19	363 mM	Greagent, Shanghai, China
Na ₂ SO ₄	3.55	25.0 mM	Greagent, Shanghai, China
KCl	0.599	8.04 mM	Greagent, Shanghai, China
NaHCO ₃	0.174	2.07 mM	RichJoint, Shanghai, China
KBr	0.0863	725 µM	RichJoint, Shanghai, China
H ₃ BO ₃	0.0230	372 µM	RichJoint, Shanghai, China
NaF	0.0028	65.7 µM	Macklin, Shanghai, China
Salt solution II –hydrated salts			
MgCl ₂ ·6H ₂ O	9.592	41.2 mM	Greagent, Shanghai, China
CaCl ₂ ·2H ₂ O	1.344	9.14 mM	Greagent, Shanghai, China
SrCl ₂ ·6H ₂ O	0.0218	82 µM	Greagent, Shanghai, China

Table S2. Silicon-free f/2 media [2].

Component	Stock Solution [g L ⁻¹]	Final concentration in media	Supplier
Major nutrient I-nitrate [1 mL/L]			
NaNO ₃	75	882 µM	Greagent, Shanghai, China
Major nutrient II-phosphate [1 mL L⁻¹]			
NaH ₂ PO ₄ ·H ₂ O	5	36.2 µM	Greagent, Shanghai, China
Metal stock I-iron [1 mL L⁻¹]			
FeCl ₃ ·6H ₂ O	3.15	11.7 µM	Greagent, Shanghai, China
Na ₂ EDTA·2H ₂ O	4.36	11.7 µM	Greagent, Shanghai, China
Metal stock II-trace metals [1 mL L⁻¹ add to Mtal stock I]			
CuSO ₄ ·5H ₂ O	9.8	0.0393 µM	Greagent, Shanghai, China
ZnSO ₄ ·7H ₂ O	22	0.0765 µM	Greagent, Shanghai, China
CoCl ₂ ·6H ₂ O	10	0.042 µM	Greagent, Shanghai, China
MnCl ₂ ·4H ₂ O	180	0.91 µM	Greagent, Shanghai, China

Na ₂ MoO ₄ ·2H ₂ O	6.3	0.026 μM	Greagent, Shanghai, China
Vitamin stock [1 mL L⁻¹]			
Thiamine-HCl	0.1	297 nM	RichJoint, Shanghai, China
Biotin	0.002	4.09 nM	RichJoint, Shanghai, China
B12	0.001	1.47 nM	RichJoint, Shanghai, China

Table S3. *P*-values of one-way ANOVA analysis of cell densities between different nutrient scenarios. Significant values were marked in bold and *.

Time	HNHP-HNLP	HNHP-LNHP	HNHP-LNLP	HNLP-LNHP	HNLP-LNLP	LNHP-LNLP
Day 0	0.656	0.746	0.856	0.902	0.790	0.886
Day 1	0.885	0.287	0.914	0.351	0.971	0.334
Day 2	0.332	0.097	0.220	0.421	0.774	0.596
Day 3	0.297	0.655	0.351	0.491	0.850	0.613
Day 4	0.686	0.411	0.430	0.666	0.247	0.128
Day 5	<0.001*	0.033*	<0.001*	0.029*	0.013*	<0.001*
Day 6	0.004*	0.139	0.006*	0.045*	0.756	0.075
Day 7	0.001*	0.533	<0.001*	<0.001*	0.916	<0.001*
Day 8	0.007*	0.060*	0.002*	0.207	0.422	0.057
Day 9	0.005*	0.399	0.003*	0.001*	0.681	<0.001*

Table S4. Results of two-way ANOVAs of the effects of nitrate (N) and phosphate (P) and their dual effect on cell densities. Significant values were marked in bold and *.

Time	N		P		N+P	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
Day 0	0.002	0.967	0.049	0.83	0.186	0.678
Day 1	0.607	0.458	0.386	0.552	0.692	0.430
Day 2	2.372	0.162	0.115	0.743	1.254	0.295
Day 3	0.036	0.854	1.467	0.260	0.218	0.653
Day 4	0.073	0.793	0.819	0.392	2.243	0.173
Day 5	16.638	0.003*	61.18	<0.001*	0.197	0.669
Day 6	0.870	0.378	18.327	0.002*	1.925	0.203
Day 7	0.147	0.712	56.68	<0.001*	0.289	0.605
Day 8	4.621	0.064	16.736	0.003*	0.906	0.369
Day 9	0.108	0.751	40.589	<0.001*	0.867	0.379

Table S5. Exponential growth rates (μ, d⁻¹) of *Heterosigma akashiwo* from references.

Salinity (PSU)	Temp (°C)	Light (μmol m ⁻² s ⁻¹)	Nitrate (μmol L ⁻¹)	Phosphate (μmol L ⁻¹)	Grow rate (d ⁻¹)	References
31	20	200	30	1	0.46±0.05	Present study
31	20	200	30	0.5	0.37±0.02	
31	20	200	15	1	0.44±0.02	
31	20	200	15	0.5	0.34±0.01	

30	10	60			0.36±0.068	
30	15	60			0.385±0.05	
30	20	60			0.55±0.24	
30	25	60			1.01±0.02	
30	30	60	PES medium		0.74±0.10	[3]
30	25	10			0.41±0.001	
30	25	50			0.01±0.082	
30	25	100			1.14±0.013	
30	25	150			0.68±0.068	
30	25	200			0.391±0.011	
20	24	60	NA	NA	0.66±0.03	
25	24	60	NA	NA	0.68±0.02	
30	24	60	NA	NA	0.63±0.03	[4]
35	24	60	NA	NA	0.60±0.03	
40	24	60	NA	NA	0.48±0.02	
31	20	60	882	36	0.35±0.05	[5]
30	20	75	882	36	0.80	
30	20	20	882	36	0.15	
30	20	70	882	36	0.53	
30	20	100	882	36	0.78	
30	20	200	882	36	0.81	
30	20	600	882	36	0.85	
30	20	100	1	36	0.54	
30	20	100	5	36	0.64	
30	20	100	10	36	0.65	
30	20	100	50	36	0.68	[6]
30	20	100	100	36	0.71	
30	20	100	200	36	0.68	
30	20	100	882	0.1	0.21	
30	20	100	882	0.3	0.55	
30	20	100	882	0.6	0.66	
30	20	100	882	3	0.78	
30	20	100	882	6	0.77	
30	20	100	882	20	0.78	
31	15	40	50	36.2	0.82±0.04	
31	15	110	50	36.2	0.46±0.08	
31	15	25	882	36.2	0.25±0.10	
31	15	50	882	36.2	0.44±0.05	
31	15	80	882	36.2	0.80±0.05	[7]
31	15	110	882	36.2	0.90±0.05	
31	15	140	882	36.2	0.87±0.10	
31	15	175	882	36.2	0.84±0.10	
31	15	200	882	36.2	0.85±0.12	
23	20	100	882	36.2	0.55±0.08	[8]

23	24	100	882	36.2	0.72±0.05	
5	25	62	882	36.2	0.48±0.03	[9]
10	25	62	882	36.2	0.57±0.03	
15	25	62	882	36.2	0.76±0.03	
20	25	62	882	36.2	0.76±0.04	
25	25	62	882	36.2	0.86±0.04	
30	25	62	882	36.2	0.63±0.02	
32	15	100	882	36	0.70±0.10	[10]
30	20	70	100	7	0.33±0.05	[11]
30	20	70	882	36	0.44±0.06	[12]
35	17	100	882	36	0.57	[13]
20	17	100	882	36	0.47	
35	23	100	882	36	0.46	
20	23	100	882	36	0.32	
31	20	45	0	36	0.30±0.10	[14]
31	20	45	128	36	0.40±0.20	
31	20	45	512	36	0.42±0.20	
31	20	45	883	0	0.28±0.07	
31	20	45	883	8	0.33±0.20	
31	20	45	883	32	0.35±0.20	
25	15	75	882	36.2	0.46±0.02	[15]
25	15	75	882	36.2	0.52±0.02	
30	24	350	3	36.2	1.00±0.05	[16]
30	24	350	10	36.2	1.10±0.10	
30	24	350	25	36.2	1.30±0.10	
30	24	350	50	36.2	1.15±0.05	
30	24	350	882	36.2	1.20±0.05	
30	24	175	882	36.2	1.10±0.05	
20	25	80	220	36.2	0.25±0.05	[17]
32	20	80	882	36	0.51	[18]
32	18	100	576	36.2	0.47±0.12	[19]
32	18	100	5	36.2	0.36±0.02	
32	18	100	882	0.2	0.37±0.12	
32	18	100	88.2	2.4	0.47±0.05	[20]
32	18	100	882	36.2	0.61±0.09	[21]
32	18	100	882	36.2	0.63±0.04	
30	20	70	880	32	0.52	[22]

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