

**This is a self-archived version of an original article. This version may differ from the original in pagination and typographic details.**

**Author(s):** Stevčić, Čedomir; Pulkkinen, Katja; Pirhonen, Juhani

**Title:** Screening of microalgae and LED grow light spectra for effective removal of dissolved nutrients from cold-water recirculating aquaculture system (RAS) wastewater

**Year:** 2019

**Version:** Accepted version (Final draft)

**Copyright:** © 2019 Elsevier B.V.

**Rights:** CC BY-NC-ND 4.0

**Rights url:** <https://creativecommons.org/licenses/by-nc-nd/4.0/>

**Please cite the original version:**

Stevčić, Č., Pulkkinen, K., & Pirhonen, J. (2019). Screening of microalgae and LED grow light spectra for effective removal of dissolved nutrients from cold-water recirculating aquaculture system (RAS) wastewater. *Algal Research*, 44, Article 101681.  
<https://doi.org/10.1016/j.algal.2019.101681>

## SUPPLEMENTARY DATA

Screening of microalgae and LED grow light spectra for effective removal of dissolved nutrients from cold-water recirculating aquaculture system (RAS) wastewater

Čedomir Stevčić \*, Katja Pulkkinen, Juhani Pirhonen

University of Jyväskylä, Department of Biological and Environmental Science, P.O. Box 35, FI-40014 University of Jyväskylä, Finland

\* Corresponding author.

E-mail address: [cedomir.stevcic@jyu.fi](mailto:cedomir.stevcic@jyu.fi) (C. Stevčić).

Table S.1. Differences in growth responses or nutrient removal among six species of green microalgae (Microalga) in two different media (Medium), tested with two-way ANOVA (upper; Run as a random factor) and Kruskal-Wallis H tests (lower). df: degree of freedom; MS: mean squares; F: value of the F statistic; p: significance level; N: number of samples;  $\chi^2$ : value of the chi-square statistic. Den: density; SGR: specific growth rate;  $R_N$ : removal rate of  $\text{NO}_3\text{-N}$ ;  $R_P$ : removal rate of  $\text{PO}_4\text{-P}$ ;  $V_P$ : cell uptake rate of  $\text{PO}_4\text{-P}$ ;  $N\%$ : percentage of  $\text{NO}_3\text{-N}$  removal;  $V_N$ : cell uptake rate of  $\text{NO}_3\text{-N}$ ;  $P\%$ : percentage of  $\text{PO}_4\text{-P}$  removal. Remarked in bold, statistically significant values ( $p < 0.05$ ).

Variable	Source	df	MS	F	p
<i>Den</i>	Microalga	5	16.783	35.406	<b>0.000</b>
	Medium	1	0.368	0.776	0.388
	Microalga*Medium	5	0.398	0.84	0.536
	Run	2	0.874	1.845	0.182
	Error	22	0.474		
<i>SGR</i>	Microalga	5	0.037	2.384	0.072
	Medium	1	0.034	2.187	0.153
	Microalga*Medium	5	0.015	1.007	0.437
	Run	2	0.021	1.348	0.280
	Error	22	0.015		
$R_N$	Microalga	5	2.998	9.098	<b>0.000</b>
	Medium	1	0.117	0.354	0.558
	Microalga*Medium	5	0.403	1.222	0.332
	Run	2	4.590	13.931	<b>0.000</b>
	Error	22	0.330		
$R_P$	Microalga	5	0.001	0.733	0.606
	Medium	1	0.502	276.685	<b>0.000</b>
	Microalga*Medium	5	0.001	0.490	0.780
	Run	2	0.035	19.569	<b>0.000</b>
	Error	22	0.002		
$V_P$	Microalga	5	5.807	131.212	<b>0.000</b>
	Medium	1	0.336	7.602	<b>0.012</b>
	Microalga*Medium	5	0.095	2.138	0.099
	Run	2	0.013	0.291	0.750
	Error	22	0.044		
$N\%$	Microalga	5	1572.128	10.439	<b>0.000</b>
	Medium	1	835.114	5.545	<b>0.028</b>
	Microalga*Medium	5	154.398	1.025	0.427
	Run	2	73.205	0.486	0.621
	Error	22	150.597		
Variable	Source	N	$\chi^2$	df	p
$V_N$	Microalga	36	32.545	5	<b>0.000</b>
	Medium	36	0.042	1	0.837
$P\%$	Microalga	36	8.068	5	0.153

Table S.2. Differences in growth responses or nutrient removal among four species of non-green microalgae (Microalga) in two different media (Medium), tested with two-way ANOVA (upper; Run as a random factor) and Kruskal-Wallis H tests (lower). df: degree of freedom; MS: mean squares; F: value of the F statistic; p: significance level; N: number of samples;  $\chi^2$ : value of the chi-square statistic. SGR: specific growth rate;  $R_N$ : removal rate of  $\text{NO}_3\text{-N}$ ;  $R_P$ : removal rate of  $\text{PO}_4\text{-P}$ ;  $V_N$ : cell uptake rate of  $\text{NO}_3\text{-N}$ ; N%: percentage of  $\text{NO}_3\text{-N}$  removal; P%: percentage of  $\text{PO}_4\text{-P}$  removal; Den: density;  $V_P$ : cell uptake rate of  $\text{PO}_4\text{-P}$ . Remarkd in bold, statistically significant values ( $p < 0.05$ ).

Variable	Source	df	MS	F	p
SGR	Microalga	3	0.032	3.838	<b>0.034</b>
	Medium	1	0.039	4.681	<b>0.048</b>
	Microalga*Medium	3	0.030	3.589	<b>0.041</b>
	Run	2	0.007	0.821	0.460
	Error	14	0.008		
$R_N$	Microalga	3	0.221	8.272	<b>0.002</b>
	Medium	1	0.010	0.360	0.558
	Microalga*Medium	3	0.010	0.370	0.776
	Run	2	0.034	1.263	0.313
	Error	14	0.027		
$R_P$	Microalga	3	0.003	27.987	<b>0.000</b>
	Medium	1	0.004	38.884	<b>0.000</b>
	Microalga*Medium	3	0.001	8.241	<b>0.002</b>
	Run	2	0.000	0.769	0.482
	Error	14	0.000		
$V_N$	Microalga	3	12.368	0.049	0.985
	Medium	1	782.498	3.094	0.100
	Microalga*Medium	3	78.611	0.311	0.817
	Run	2	828.115	3.275	0.068
	Error	14	252.884		
N%	Microalga	3	747.256	5.996	<b>0.008</b>
	Medium	1	4.735	0.038	0.848
	Microalga*Medium	3	44.188	0.355	0.787
	Run	2	111.437	0.894	0.431
	Error	14	124.632		
P%	Microalga	3	1282.815	11.878	<b>0.000</b>
	Medium	1	123.261	1.141	0.303
	Microalga*Medium	3	92.554	0.857	0.486
	Run	2	169.254	1.567	0.243
	Error	14	108.002		
Variable	Source	N	$\chi^2$	df	p
Den	Microalga	24	16.411	3	<b>0.001</b>
	Medium	24	49.500	1	0.186
$V_P$	Microalga	24	11.028	3	<b>0.012</b>
	Medium	24	56.000	1	0.347

Table S.3. Differences in growth responses or nutrient removal among three species of green microalgae (Microalga) on three different LED grow light spectra (Spectrum), tested with two-way ANOVA (Run as a random factor). df: degree of freedom; MS: mean squares; F: value of the F statistic; p: significance level. Den: density; SGR: specific growth rate; DW: dry weight; Chl a: chlorophyll-a concentration;  $R_N$ : removal rate of  $NO_3-N$ ;  $R_P$ : removal rate of  $PO_4-P$ ;  $V_N$ : cell uptake rate of  $NO_3-N$ ;  $V_P$ : cell uptake rate of  $PO_4-P$ ; N%: percentage of  $NO_3-N$ ; P%: percentage of  $PO_4-P$  removal. Remarked in bold, statistically significant values ( $p < 0.05$ ).

Variable	Source	df	MS	F	p
<i>Den</i>	Microalga	2	119.577	55.312	<b>0.000</b>
	Spectrum	2	0.522	0.241	0.788
	Microalga*Spectrum	4	0.305	0.141	0.964
	Run	2	13.807	6.387	<b>0.009</b>
	Error	16	2.162		
<i>SGR</i>	Microalga	2	0.075	7.909	<b>0.004</b>
	Spectrum	2	0.006	0.634	0.543
	Microalga*Spectrum	4	0.000	0.048	0.995
	Run	2	0.052	5.461	<b>0.016</b>
	Error	16	0.009		
<i>DW</i>	Microalga	2	0.043	6.101	<b>0.011</b>
	Spectrum	2	0.004	0.515	0.607
	Microalga*Spectrum	4	0.002	0.290	0.880
	Run		0.089	12.568	<b>0.001</b>
	Error	16	0.007		
<i>Chl a</i>	Microalga	2	20.019	31.267	<b>0.000</b>
	Spectrum	2	0.778	1.215	0.323
	Microalga*Spectrum	4	0.184	0.287	0.882
	Run	2	70.453	110.036	<b>0.000</b>
	Error	16	0.640		
$R_N$	Microalga	2	1.456	25.021	<b>0.000</b>
	Spectrum	2	0.043	0.732	0.496
	Microalga*Spectrum	4	0.037	0.641	0.641
	Run	2	8.338	143.266	<b>0.000</b>
	Error	16	0.058		
$R_P$	Microalga	2	0.027	48.902	<b>0.000</b>
	Spectrum	2	0.000	0.080	0.923
	Microalga*Spectrum	4	0.000	0.080	0.987
	Run	2	0.066	118.797	<b>0.000</b>
	Error	16	0.001		
$V_N$	Microalga	2	5.158	70.641	<b>0.000</b>
	Spectrum	2	0.022	0.304	0.742
	Microalga*Spectrum	4	0.006	0.079	0.988
	Run	2	0.119	1.627	0.227
	Error	16	0.073		
$V_P$	Microalga	2	0.009	41.583	<b>0.000</b>

	Spectrum	2	0.000	0.221	0.804
	Microalga*Spectrum	4	0.000	0.272	0.891
	Run	2	0.002	7.166	<b>0.006</b>
	Error	16	0.000		
<i>N%</i>	Microalga	2	392.461	37.779	<b>0.000</b>
	Spectrum	2	10.564	1.017	0.384
	Microalga*Spectrum	4	7.847	0.755	0.569
	Run	2	2620.119	252.220	<b>0.000</b>
	Error	16	10.388		
<i>P%</i>	Microalga	2	2929.713	501.371	<b>0.000</b>
	Spectrum	2	2.951	0.505	0.613
	Microalga*Spectrum	4	6.875	1.176	0.358
	Run	2	8.637	1.478	0.258
	Error	16	5.843		

Table S.4. Differences in growth responses or nutrient removal of *H. pluvialis* on three different LED grow light spectra, tested with one-way ANOVA and Welch ANOVA. F: value of the F statistic; df: degrees of freedom; p: significance level. Den: density; SGR: specific growth rate; DW: dry weight; Chl a: chlorophyll-a concentration;  $R_N$ : removal rate of  $\text{NO}_3\text{-N}$ ;  $R_P$ : removal rate of  $\text{PO}_4\text{-P}$ ;  $V_N$ : cell uptake rate of  $\text{NO}_3\text{-N}$ ;  $V_P$ : cell uptake rate of  $\text{PO}_4\text{-P}$ ;  $N\%$ : percentage of  $\text{NO}_3\text{-N}$ ;  $P\%$ : percentage of  $\text{PO}_4\text{-P}$  removal. Remarked in bold, statistically significant values.

Variable	Analysis	F	df1	df2	p
<i>Den</i>	ANOVA	1.5	2	6	0.301
<i>SGR</i>	ANOVA	2.4	2	6	0.167
<i>DW</i>	ANOVA	5.4	2	6	<b>0.046</b>
<i>Chl a</i>	ANOVA	15.5	2	6	<b>0.004</b>
$R_N$	Welch ANOVA	22.6	2	3.5	<b>0.010</b>
$R_P$	ANOVA	22.8	2	6	<b>0.002</b>
$V_N$	Welch ANOVA	30.2	2	3.6	<b>0.006</b>
$V_P$	ANOVA	4.2	2	6	0.073
$N\%$	Welch ANOVA	21.8	2	3.5	<b>0.011</b>
$P\%$	ANOVA	35.9	2	6	<b>0.000</b>