Biomass productivity and fatty acid accumulation of *Neochloris oleoabundans* under alkaline-saline conditions

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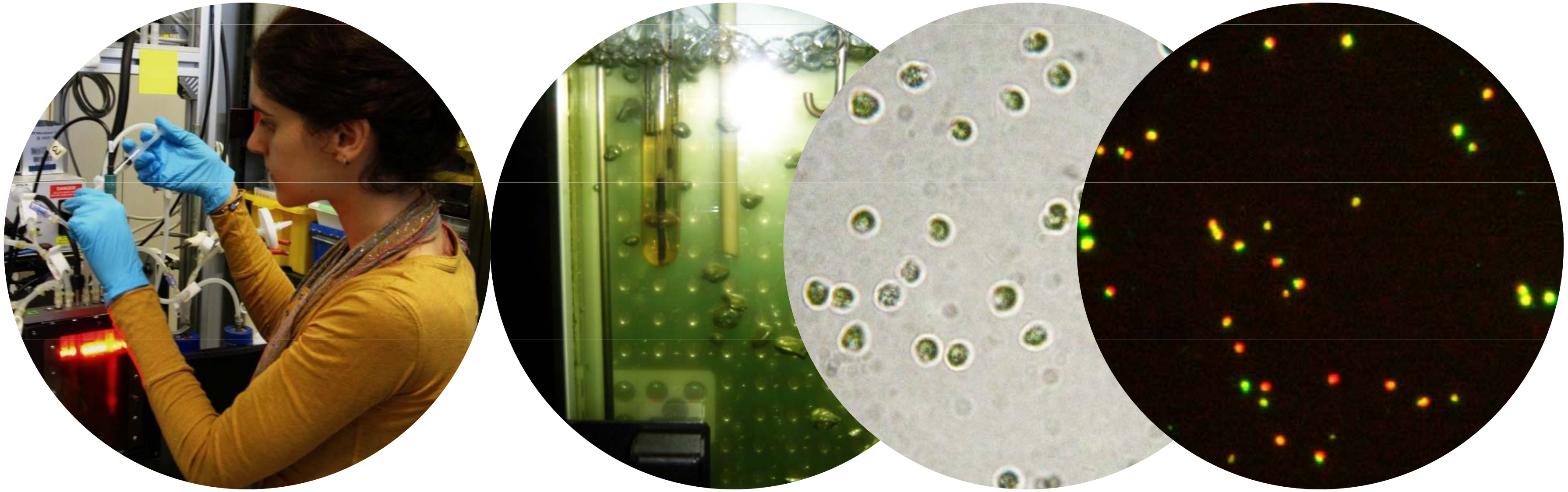

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Microalgal cultivation

Potential:

Sustainable biofuels production

Limitations in PBR´s:

Lipid yields on light energy

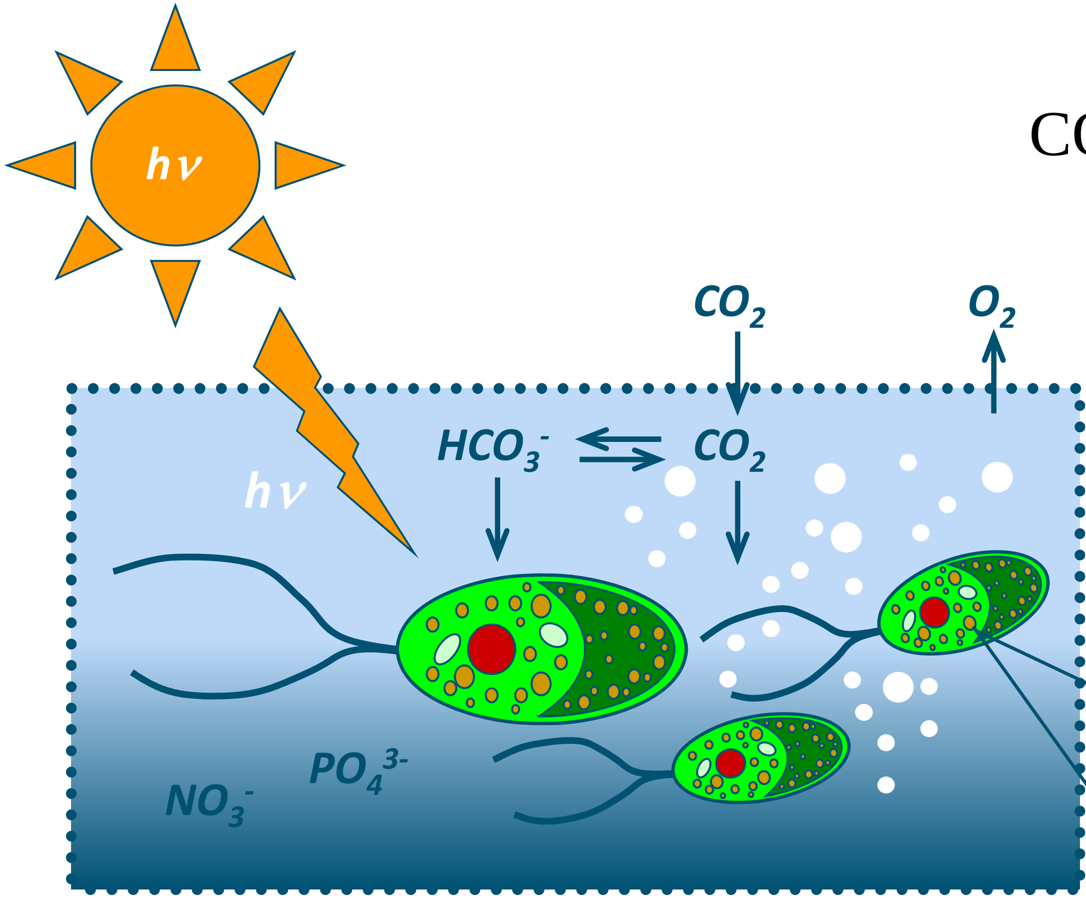
CO_2 transfer

Solution:

Maximize biomass and lipid productivities

Optimization at elevated pH?

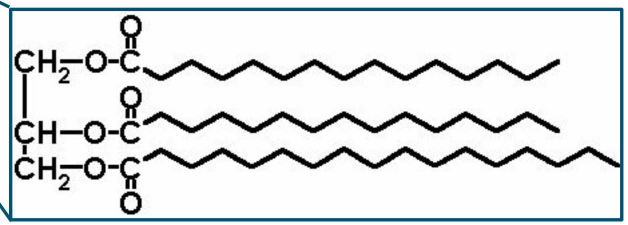
Growing lipid-rich microalgae under alkaline-saline conditions



$$CO_2 + H_2O \xleftrightarrow{0.039\ s^{-1}} HCO_3^- + H^+$$

$$CO_2 + OH^- \xleftrightarrow{10^3\ s^{-1}} HCO_3^-$$

Triacylglyceride



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How to trigger lipid accumulation in microalgae?

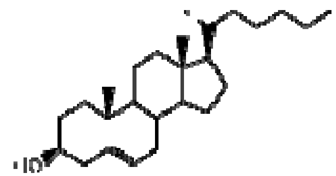
High light intensities

Sub-optimal growth temperatures

High salinity

Extreme pH

Nutrient deprivation

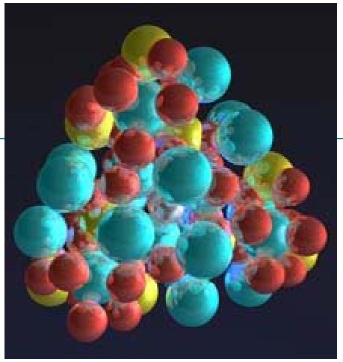


Neutral lipid

Neochloris oleoabundans

Medium design

Seawater-type media with reduced Ca^{2+} and PO_4^{3-}



$Ca_5(PO_4)_3OH$

Hydroxiapatite

pH 8.2

420 mM NaCl

10 mM HCO_3^-

pH 10

270 mM NaCl

75 mM HCO_3^-

75 mM CO_3^{2-}

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Chemostat experiments

Flat-panel PBR (FMT150)

T = 30 ºC
PFD_{in} = 440 $\mu mol_{photons}$ m^{-2} s^{-1}

pH = 8.2
Salinity = 3.0%
1% CO_2

N-replete

pH = 10
Salinity = 3.4%
0.1% CO_2

N-deplete

D = 0.6 d^{-1} – 1.9 d^{-1}

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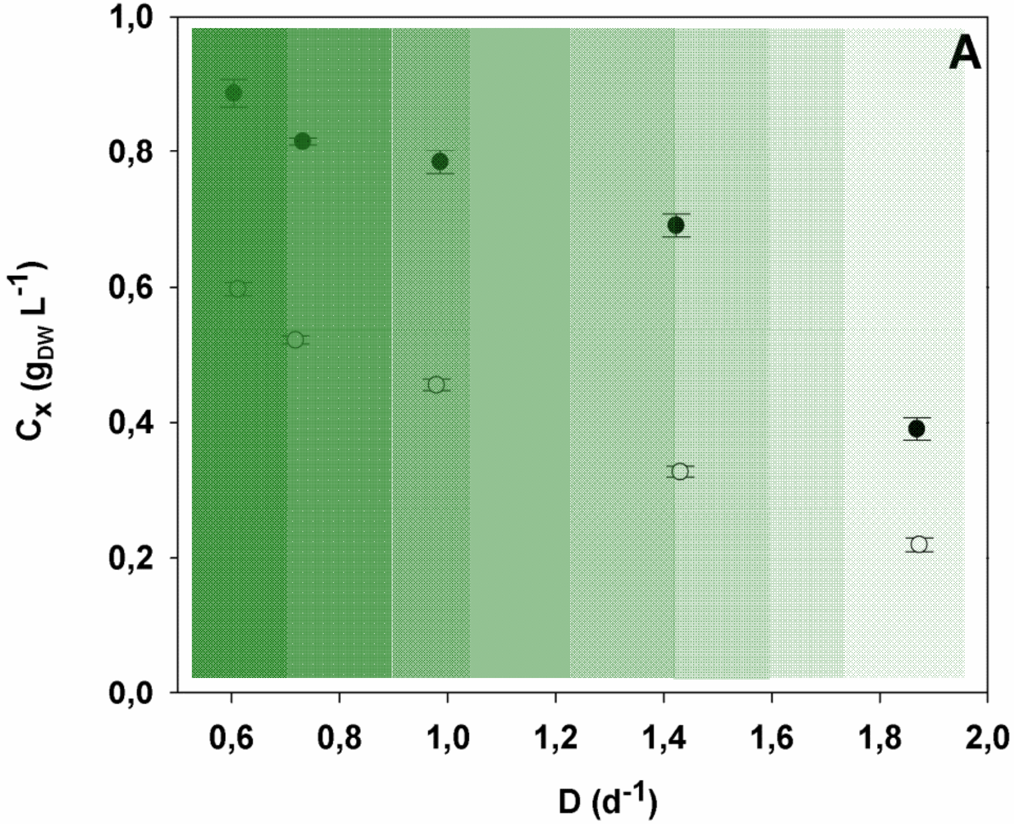
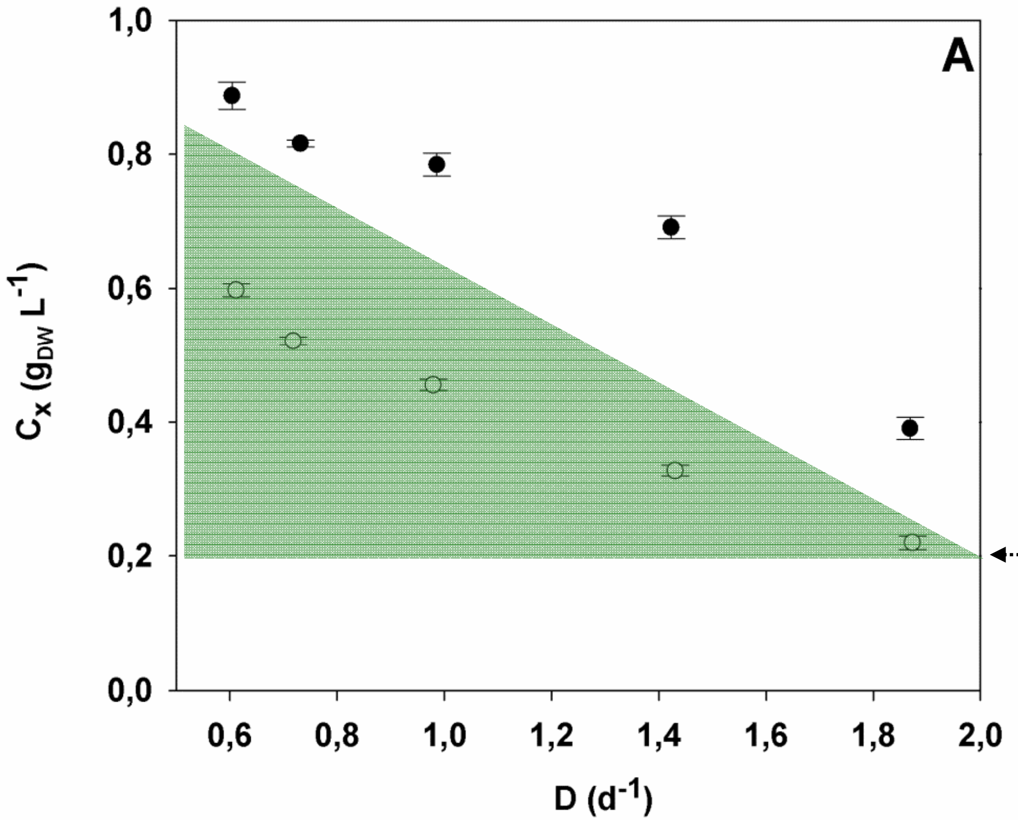
Biomass density (C_x) and productivity (P_x)



- ● pH 8.2
- ○ pH 10

C_x in steady-state set by the dilution rate (D)

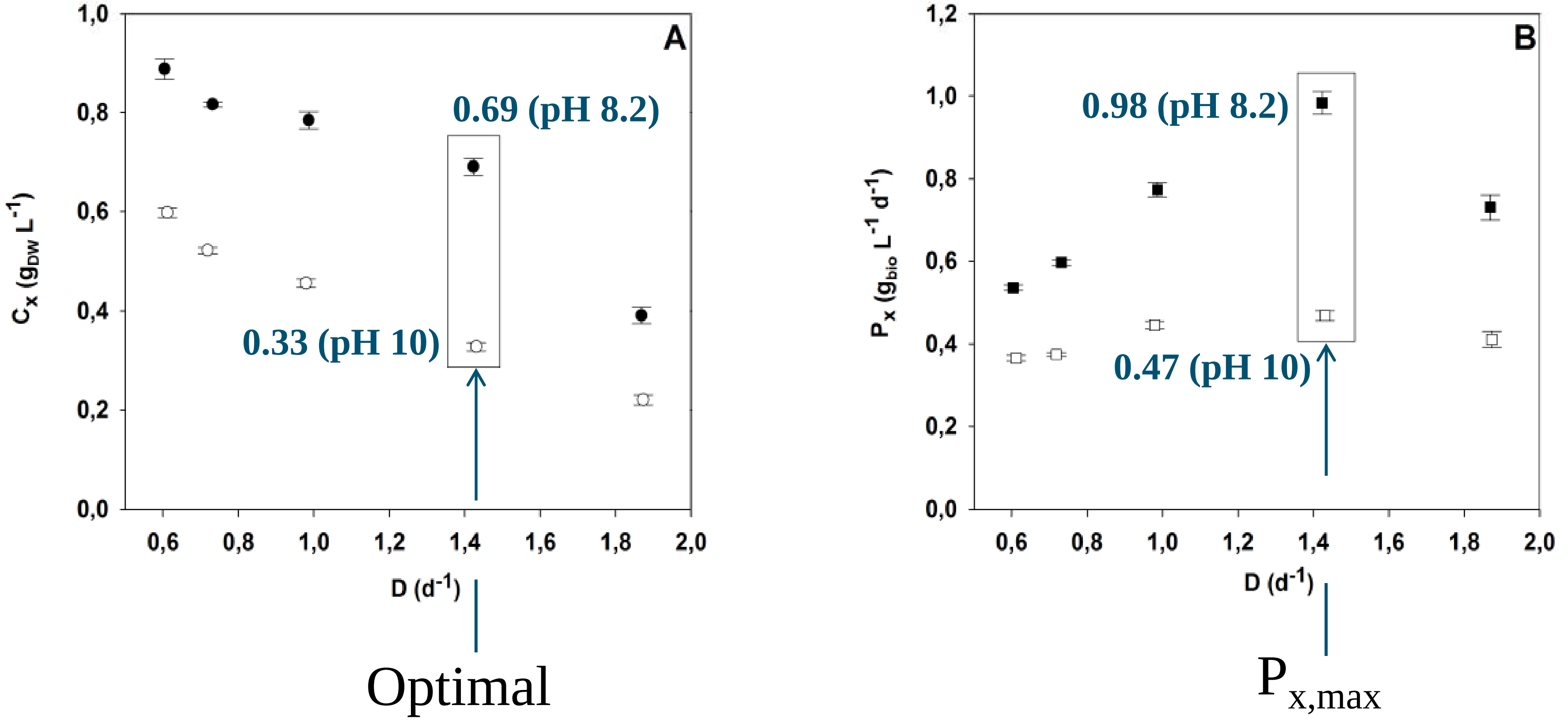
Biomass density and productivity



- pH 8.2
- pH 10

Higher light fraction exposure

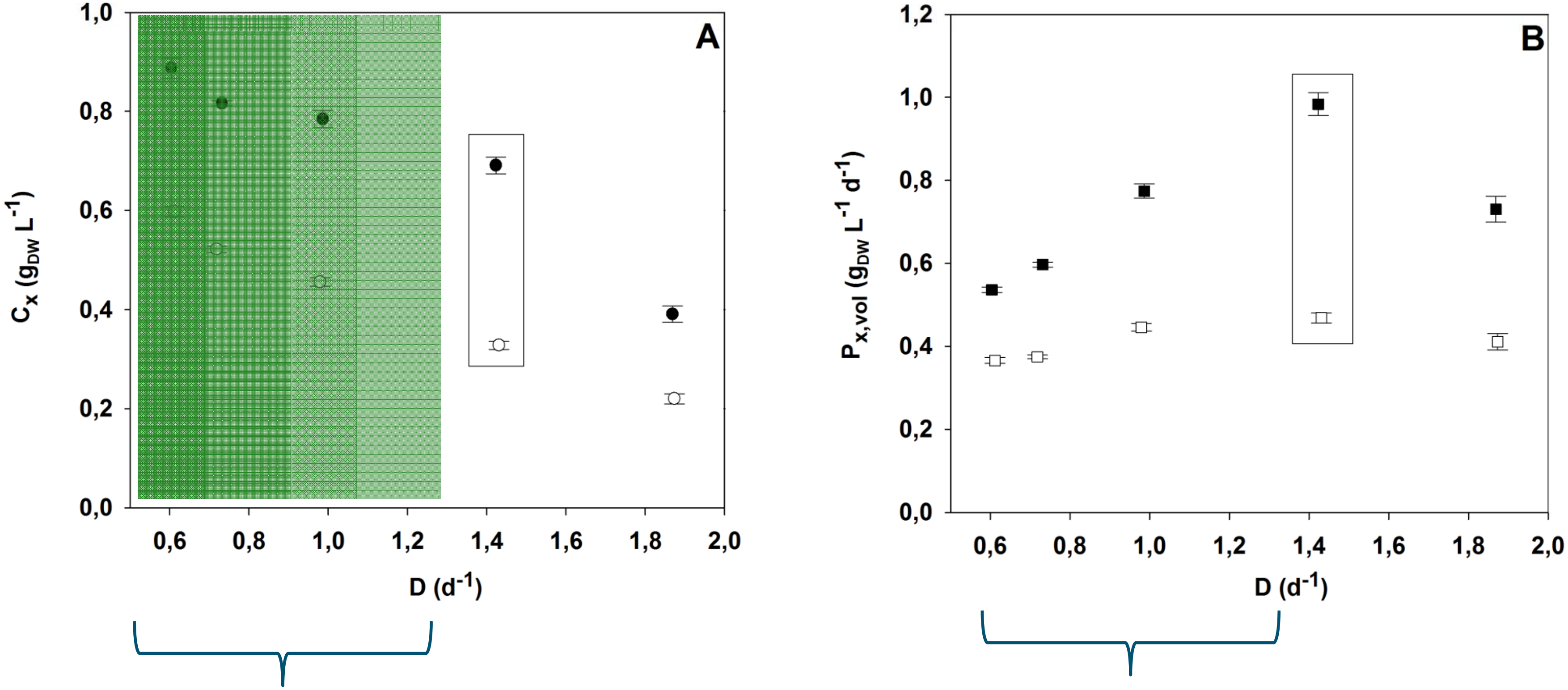
Biomass density and productivity



-Maximal light absorbed

-Minimal energy loss to maintenance

Biomass density and productivity



More energy to maintenance
Less energy for biomass formation

Sub-optimal

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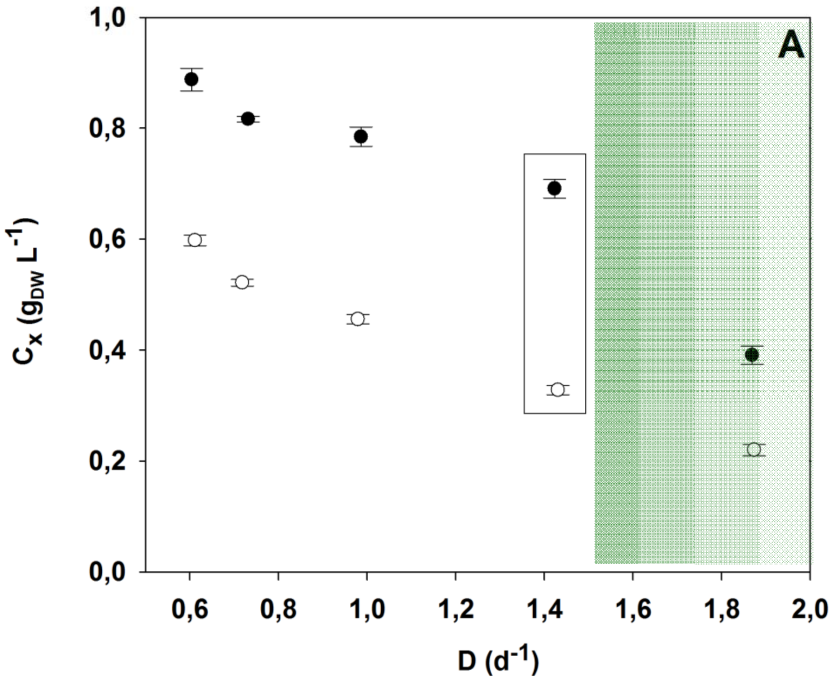
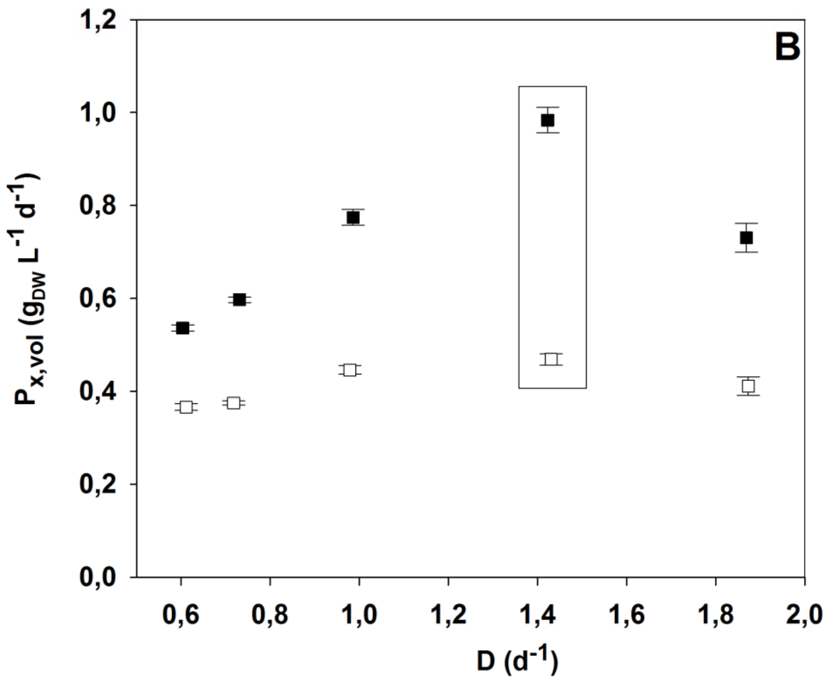
Biomass density and productivity





More light un-absorved
Less energy for biomass formation

Sub-optimal

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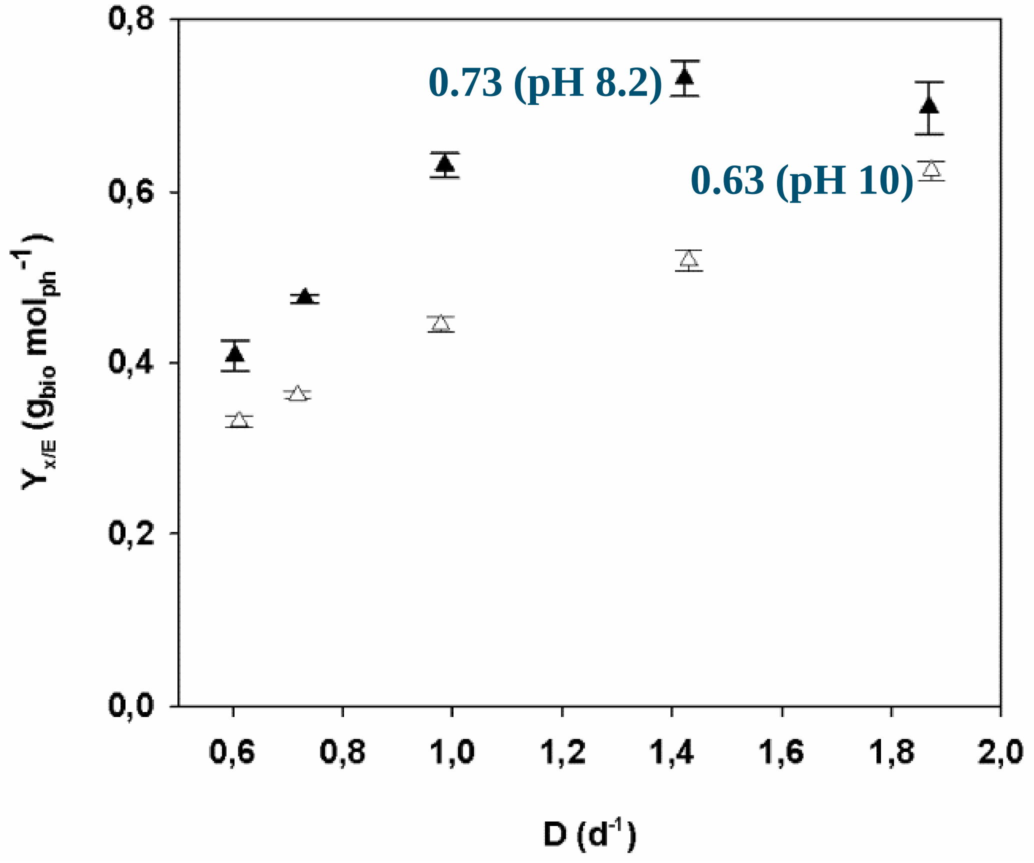
Energy requirements

Biomass yield on absorbed light energy ($Y_{x/E}$)



Fatty acid accumulation

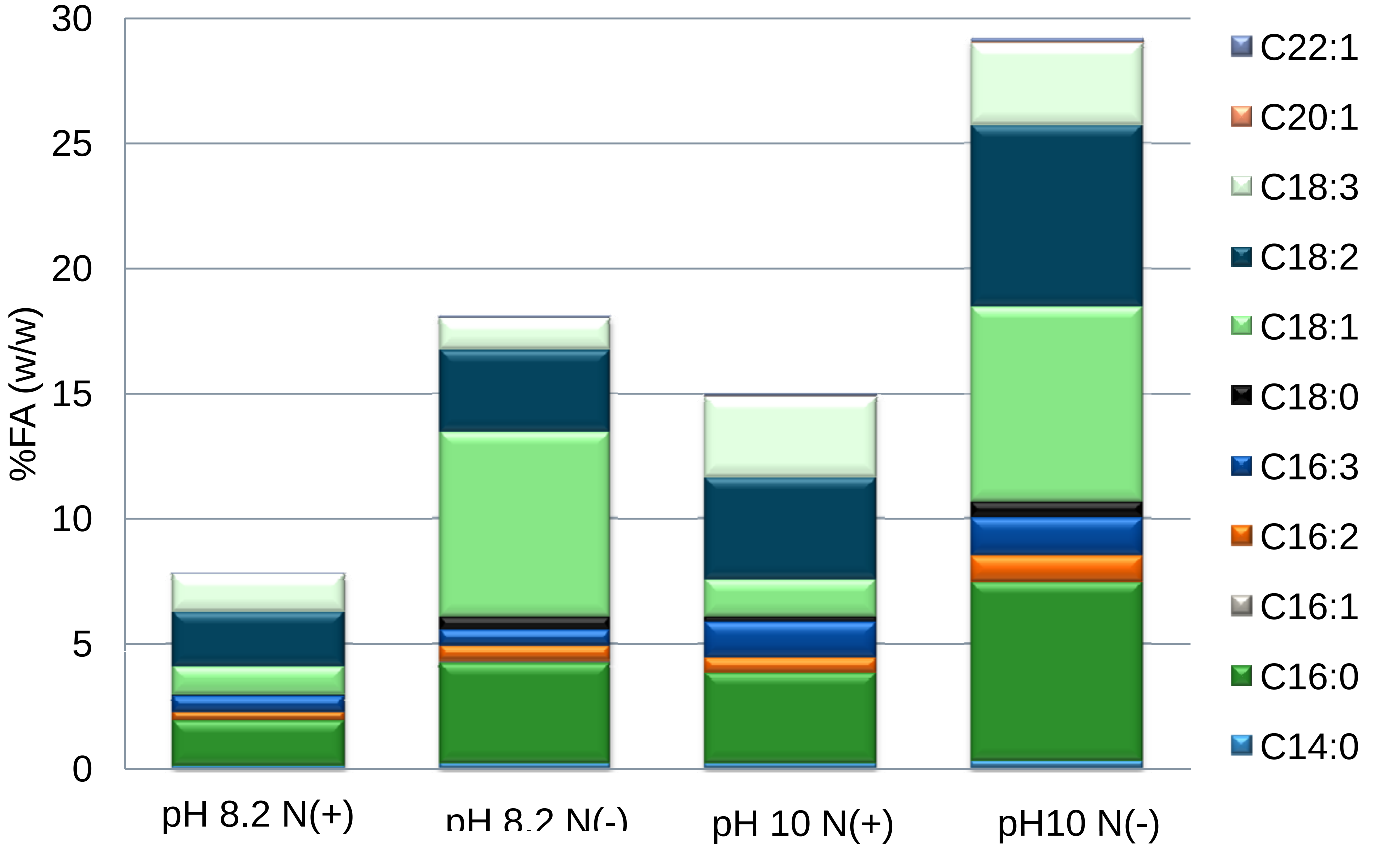
	D (d^{-1})	C_x (g_{DW} L^{-1})	$P_{x,vol}$ (g_{DW} L^{-1} d^{-1})		Total FA (% w/w)		P_{FA} (mg_{FA} L^{-1} d^{-1})	
			N(+)	N(-)	N(+)	N(-)	N(+)	N(-)
pH 8.2								
	0.6±0.02	0.89	0.54±0.02	0.33±0.01	8.74±0.17	18.18±0.01	46.85±0.93	60.52±0.05
	0.7±0.01	0.82	0.60±0.01	-	8.33±0.002	-	49.74±0.01	-
	1.0±0.01	0.78	0.77±0.02	0.32±0.01	7.53±0.01	18.52±0.01	58.30±0.11	64.56±0.03
	1.4±0.03	0.69	0.98±0.03	-	7.65	-	75.20	-
	1.9±0.004	0.39	0.73±0.03	0.37±0.02	7.13±0.01	18.11±0.01	52.07±0.09	67.54±0.02
pH 10.0								
	0.6±0.01	0.60	0.37±0.01	-	14.98±0.01	-	54.85±0.01	-
	0.7±0.004	0.52	0.38±0.004	-	14.89±0.18	-	55.84±0.67	-
	1.0±0.01	0.46	0.45±0.01	-	13.92±0.40	-	62.07±1.78	-
	1.4±0.01	0.33	0.47±0.01	-	14.19±0.03	-	66.55±0.12	-
	1.9±0.03	0.22	0.41±0.02	0.38±0.07	14.94±0.01	29.20±0.04	61.42±0.02	112.41±0.17

Fatty acids maximized at maximum biomass productivities

N-replete conditions: highest P_{FA} at reference conditions

Increase of FA content at higher pH independent from N depletion

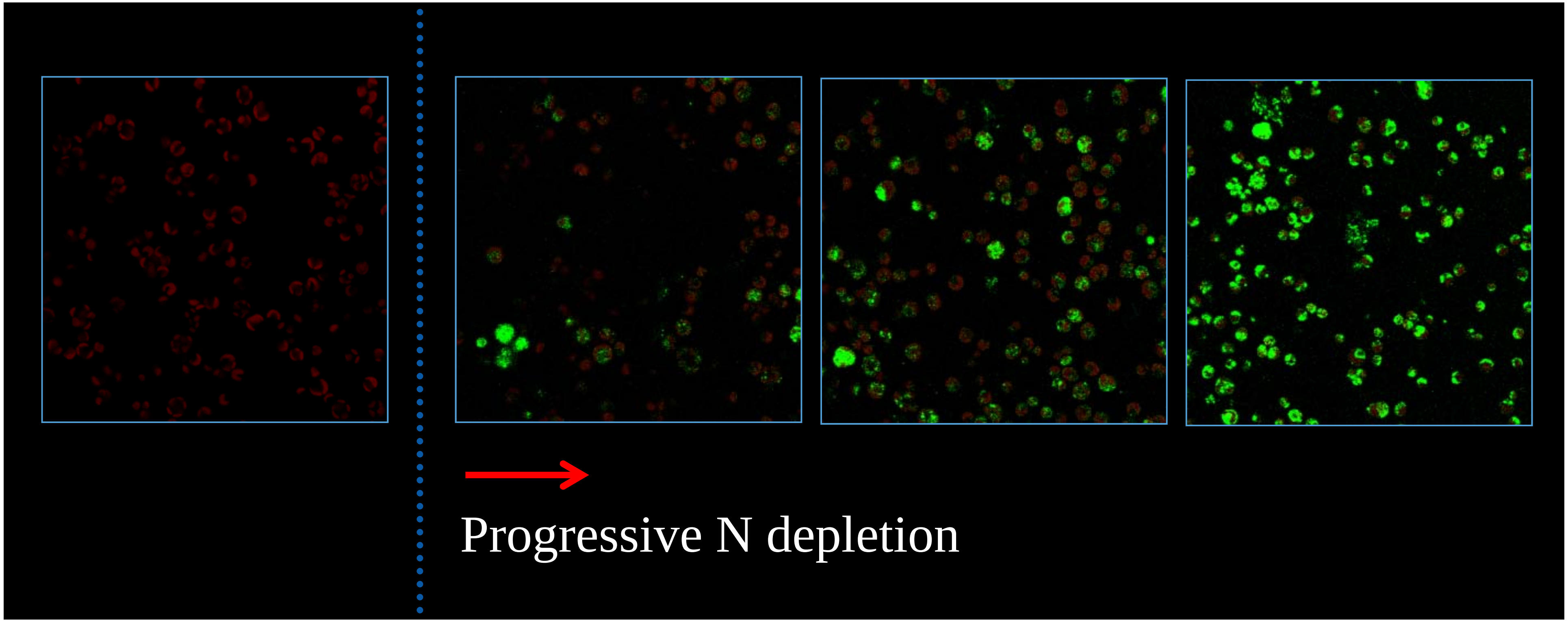
Fatty acid relative content



Increase in C16:0 and C18:1 with N depletion but also with pH

Neutral lipid accumulation

Fluorescence microscopy - Oil bodies visualization by staining with BODIPY



Conclusions

Growing *N. oleoabundans* in alkaline-saline conditions, plus nitrogen depletion, might bring an advantage in boosting lipid production for microalgal biofuels on a large-scale.

Nevertheless, nitrogen-replete biomass productivity drops substantially at elevated pH due to increased energy costs.

A two-stage process in which *N. oleoabundans* is first grown at optimal conditions and then induced to accumulate lipids using nitrogen depletion and pH upshock might be a promising strategy.

Acknowledgments

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