

SUPPORTING INFORMATION

Enhanced and selective lipid extraction from microalgae *P. tricornutum* by dimethyl carbonate and supercritical CO₂ using Deep Eutectic Solvents and Microwaves as pre-treatment

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Figure S2 Amount of lipids and TFA (determined as FAME) extracted by DMC after pre-treatment with ChCl-OA DES, varying DES-water ratio, aDES-biomass ratio and temperature.

Figure S3 Amount of lipids and TFA (determined as FAME) extracted by DMC after DES (ChCl-OA) and DES-US pre-treatments at different time and temperature conditions

Figure S4 ¹H-NMR spectra of lipid extract obtained with scCO₂ on DES (ChCl-OA) pre-treated biomass.

Single DES components pre-treatment

Method:

ChCl and OA were singularly dissolved in water to reach the same concentration present in 1ml of ChCl-OA aDES and then added to algal biomass (100 mg). The mixture was magnetically stirred for 24 hours at RT. The algal suspension was diluted with deionized water (3 mL) and then centrifuged to separate the supernatant from biomass. Algal biomass was washed two times more with deionized water (3 mL) and then freeze-dried.

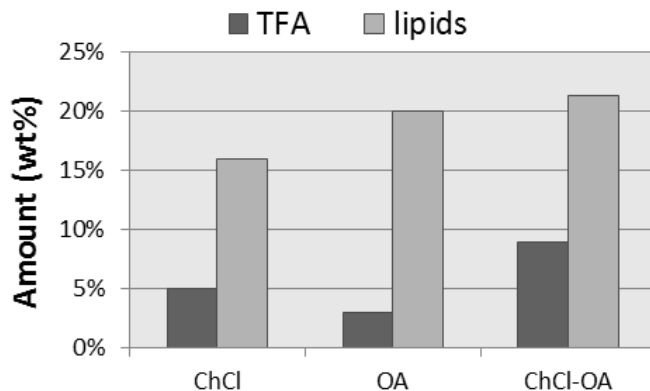
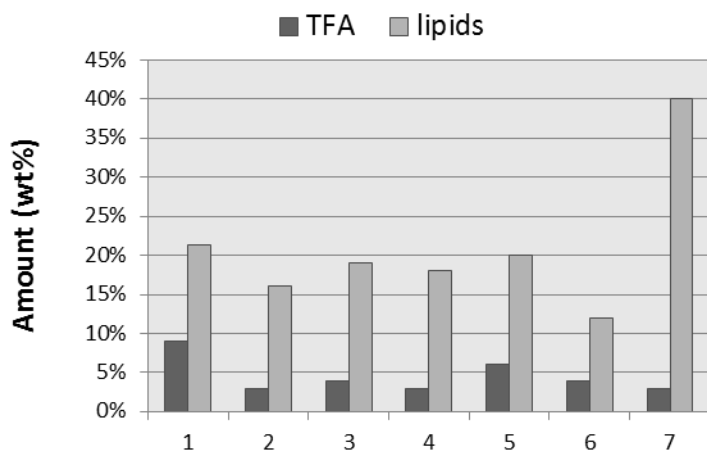


Figure S1 Amount of lipids and TFA (determined as FAME) extracted by DMC using DES and its single components (choline chloride and oxalic acid) as pre-treatments.

Study of DES pre-treatment parameters



| Entry | DES/ H ₂ O (w/w) | aDES/ bi- omass (mL/100m g) | T (°C) |
|-------|-----------------------------------|--------------------------------------|-----------|
| 1 | 3:2 | 1 | RT |
| 2 | 1:0 | 1 | RT |
| 3 | 9:1 | 1 | RT |
| 4 | 4:1 | 1 | RT |
| 5 | 3:2 | 2 | RT |
| 6 | 3:2 | 1 | 50°C |
| 7 | 3:2 | 1 | 70°C |

Figure S2 Amount of lipids and TFA (determined as FAME) extracted by DMC after pre-treatment with ChCl-OA DES, varying DES-water ratio, aDES-biomass ratio and temperature.

DES-US pre-treatment

Method:

The microalgae samples (400 mg) were added to the fresh prepared DES (4g) into a 40 mL PE centrifuge vial. The sample suspension was sonicated in a high-power bath (MG 200 TFD MF 40-80-120, Weber Ultrasonics GmbH), filled with flowing water to keep temperature constant. The vial was fixed to a rotating system to guarantee a homogeneous sonication. Based on preliminary experiments, a working frequency of 40 kHz was selected to maximize the cavitation effects on cell walls. The power was maintained constant at 200 W.

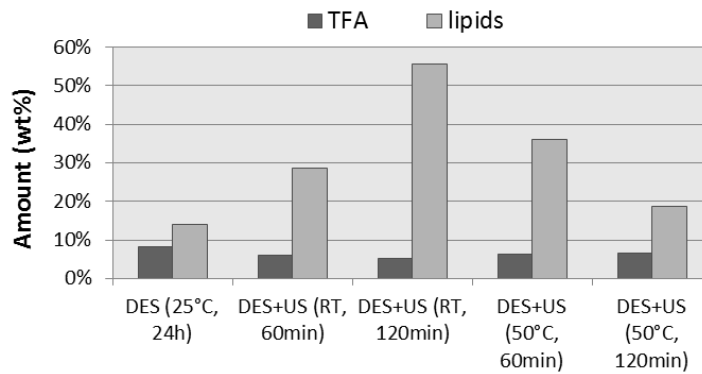
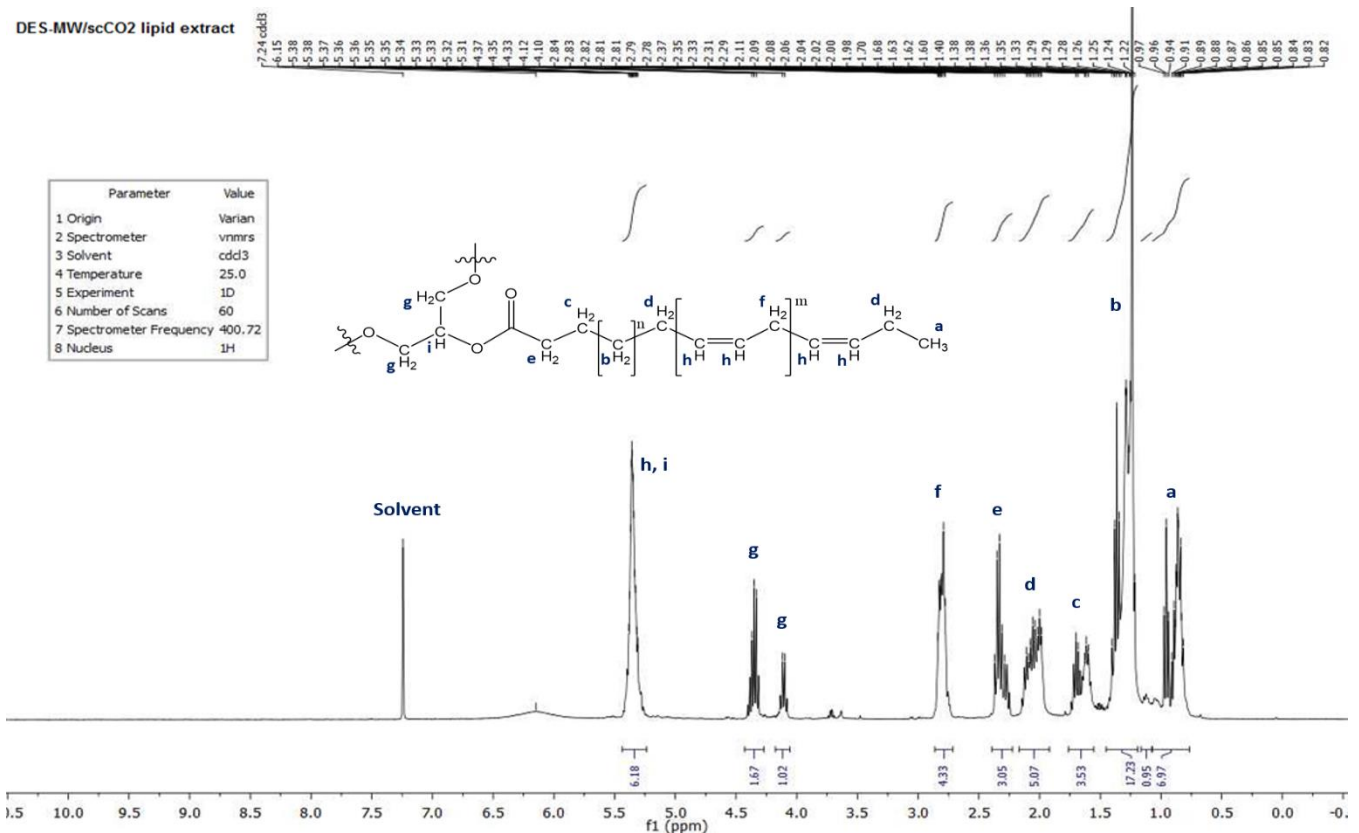


Figure S3 Amount of lipids and TFA (determined as FAME) extracted by DMC after DES (ChCl-OA) and DES-US pre-treatments at different time and temperature conditions.



¹H-NMR spectra

Figure S4 ¹H-NMR spectra of lipid extract obtained with scCO₂ on DES (ChCl-OA) pre-treated biomass. Spectra was recorded using a 5 mm probe on a Varian Mercury 400 spectrometer in CDCl₃. Signals could be mostly attributed to a mixture of triglycerides (n≥0, m≥0).