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Characteristics of lipids body and extracellular matrix in hydrocarbon-rich microalga *Botryococcus braunii* race A and race B strains

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Background

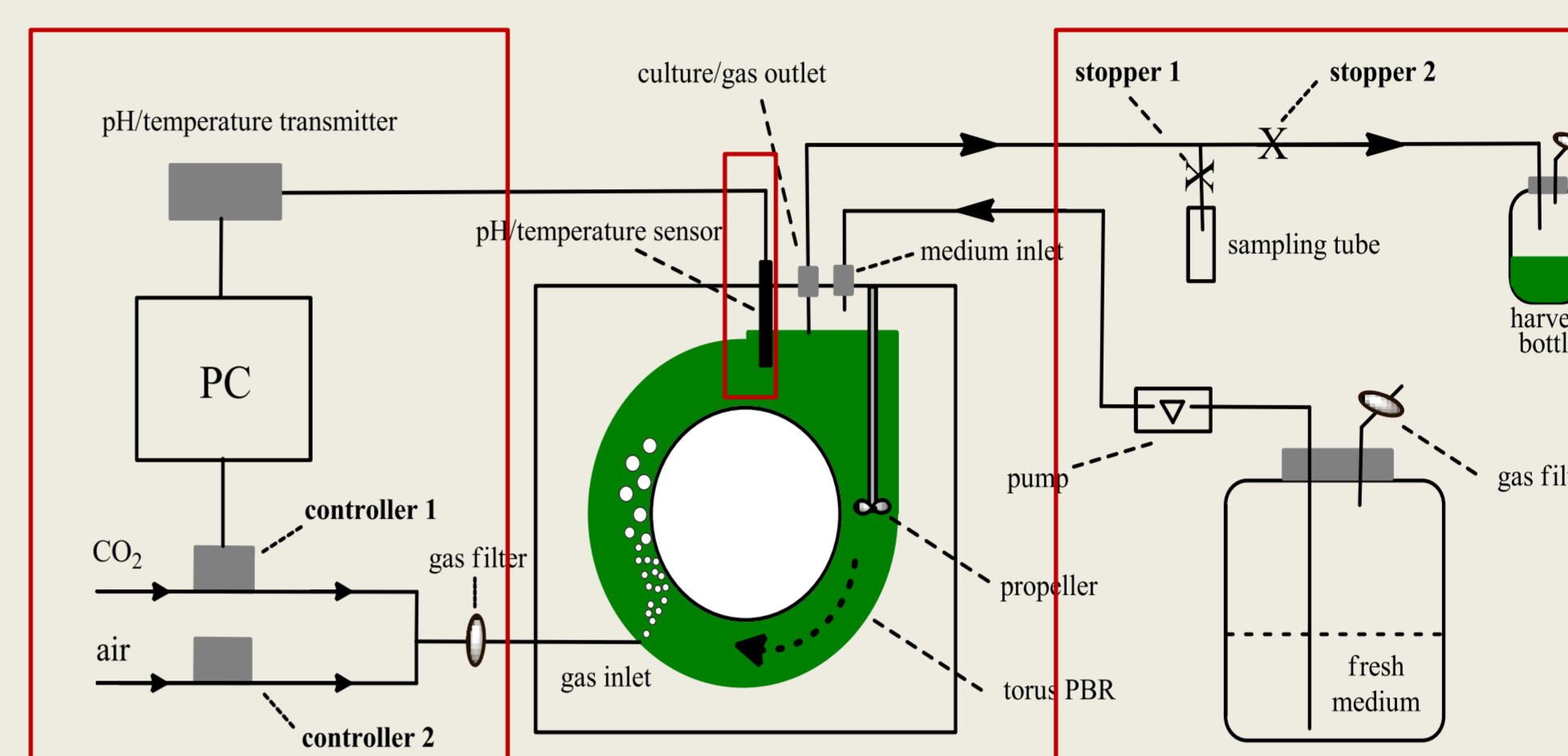
- *Botryococcus braunii* Kützing :
 - ◆ A hydrocarbon producing microalga
 - ◆ Four races (A, B, L and S) according to the chemical structures
 - ◆ Higher hydrocarbons contents in race A and B,
 - ◆ A biomass-derived alternative jet fuel

- ◆ Hydrocarbons are derived from intracellular lipids
- ◆ Hydrocarbons are accumulated extracellularly within a polymerized matrix

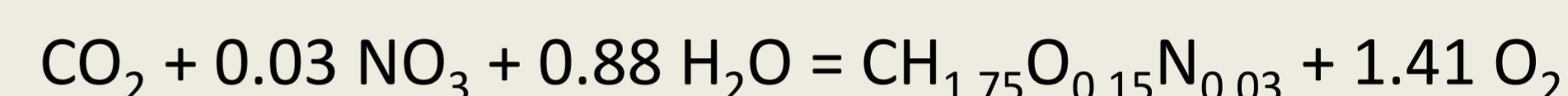
Objective

- the cells and colonies morphologies of the two strains (race A and race B) in relation with kinetic parameters of their respective cultures within a chemostat photobioreactor.

Controlled continuous cultures with two strains



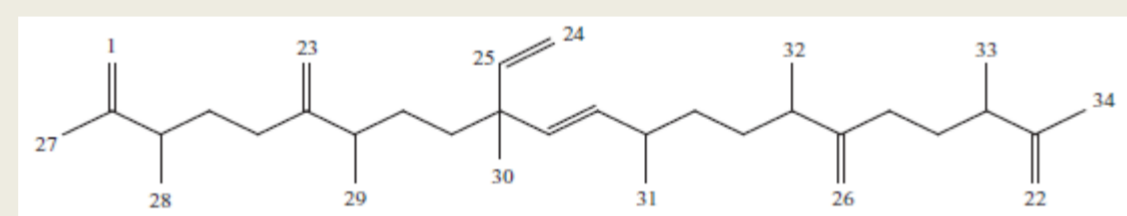
- ➔ A-race *B. braunii* strain SAG 30.81
- ➔ B-race *B. braunii* strain Bot 22
- ➔ Chemostat cultures $D_{opt} = 0.23 \text{ day}^{-1}$



Main hydrocarbons according to the strains

SAG 30.81 Alkadiens and Alkatriens

BOT22 Botryococcene $\text{C}_{34}\text{H}_{58}$



Nile Red staining → neutral lipids (yellow)
Chlorophyll → autofluorescence (red)



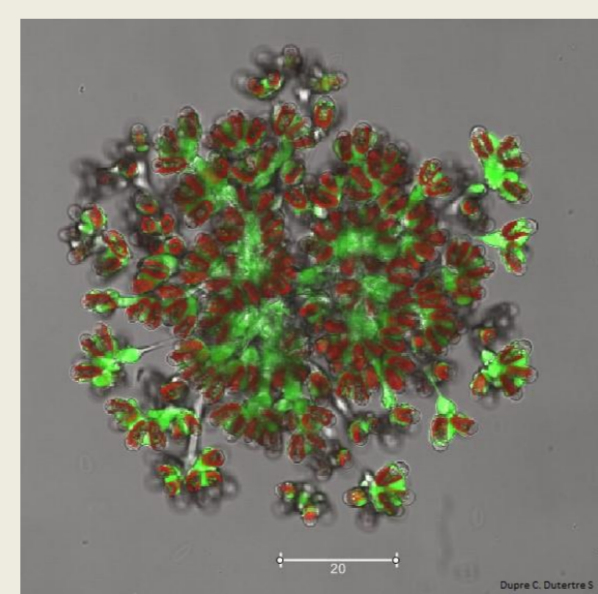
➔ Chemical analysis confirmed image analysis

Hydrocarbon race	Strain	$\text{g} \cdot \text{m}^{-2} \cdot \text{d}^{-1}$	Content (% dry weight)
Race A	SAG 30,81	0,33	5,1 ±
Race B	BOT 22	2,96	38,3 ± 3,5

No significant hydrocarbon release from cell colonies at low shear stress

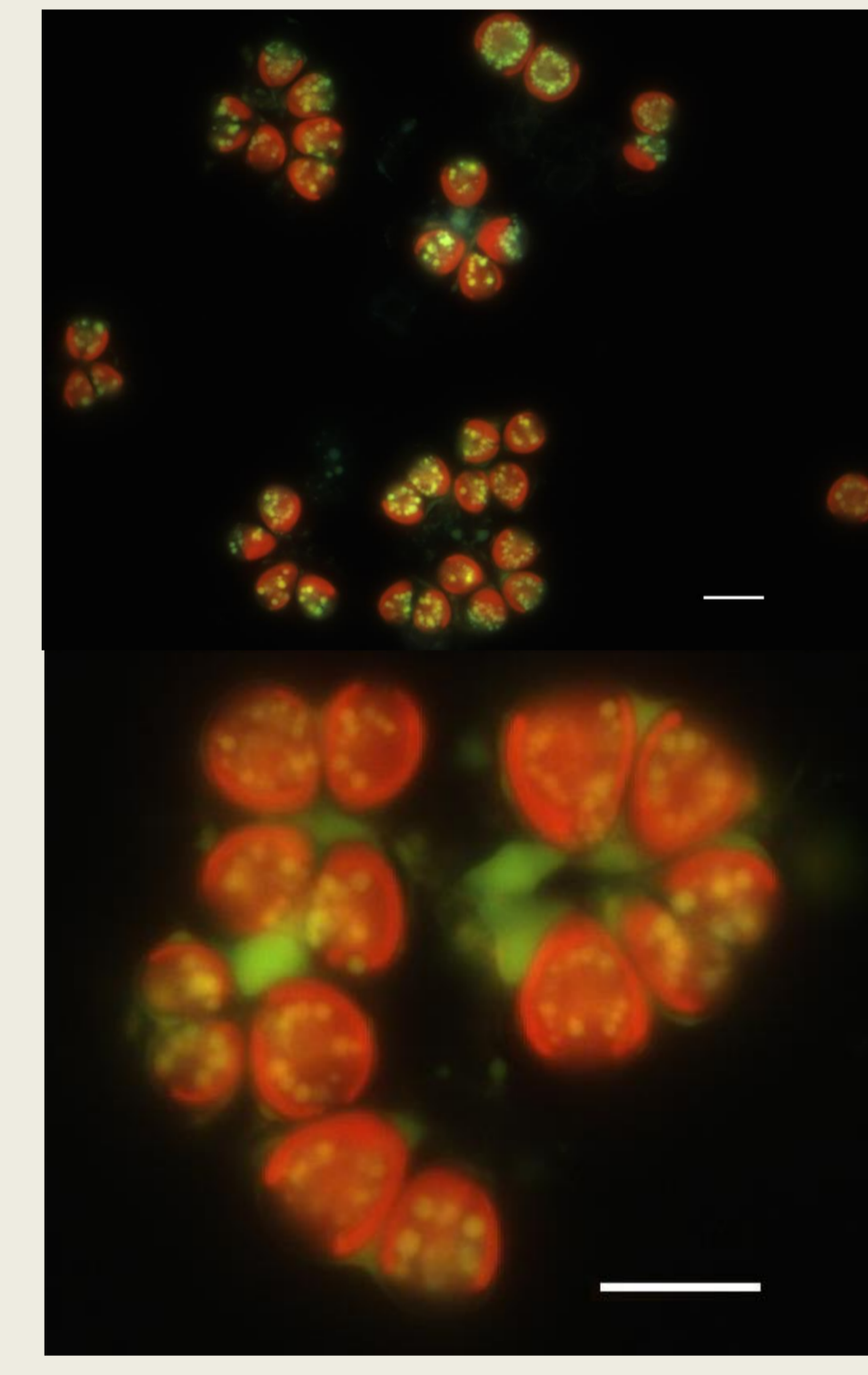
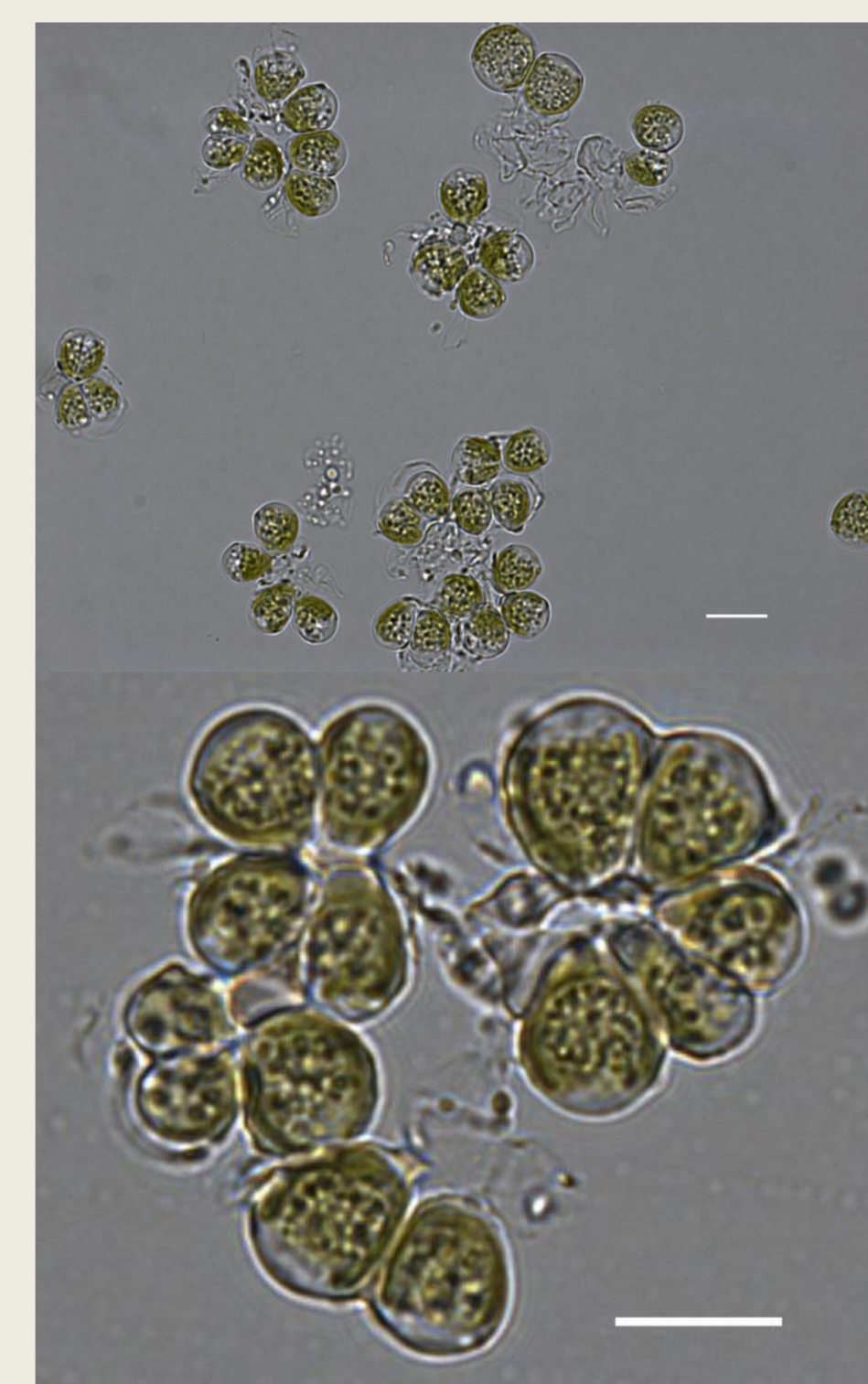


Photo Biosit



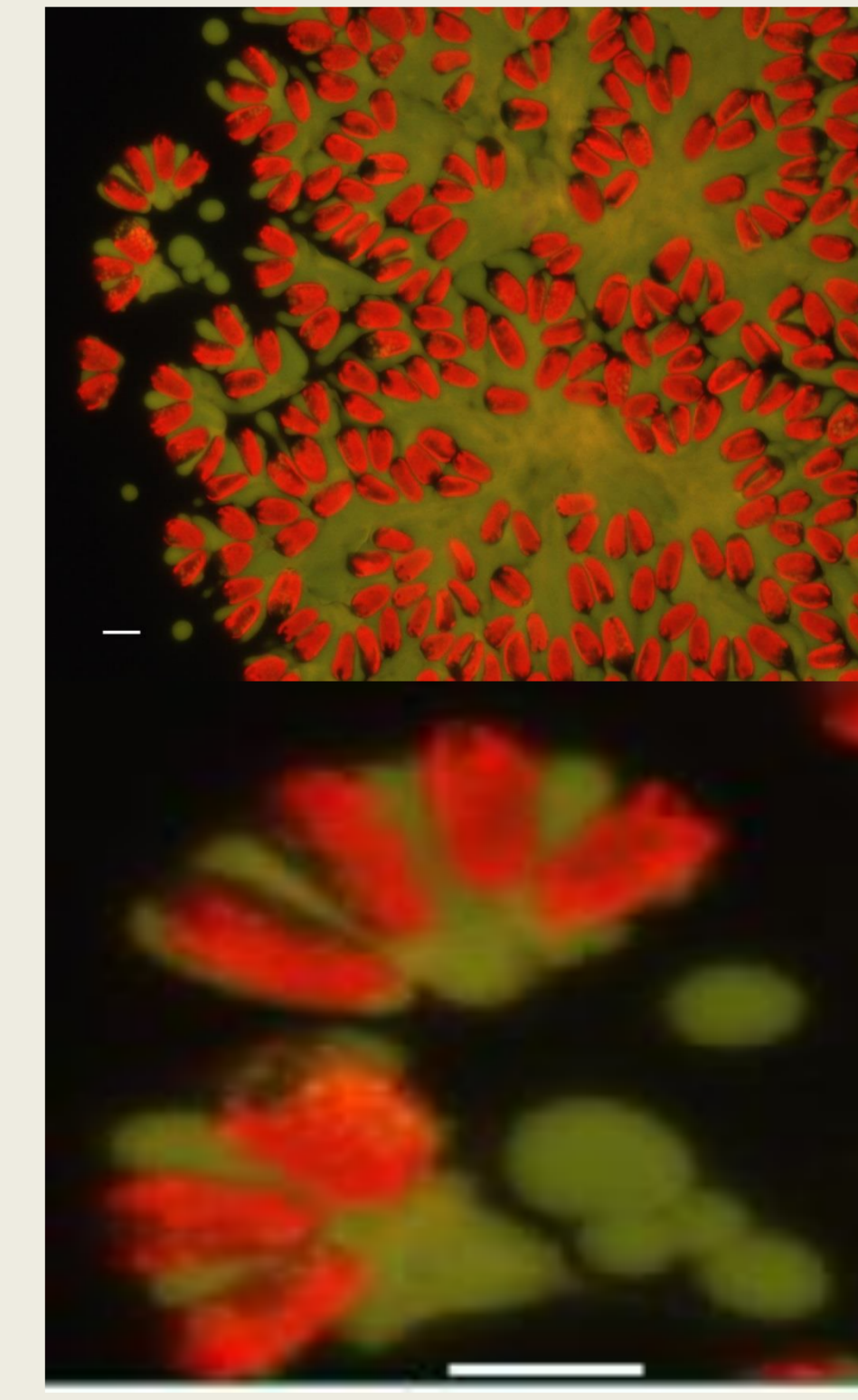
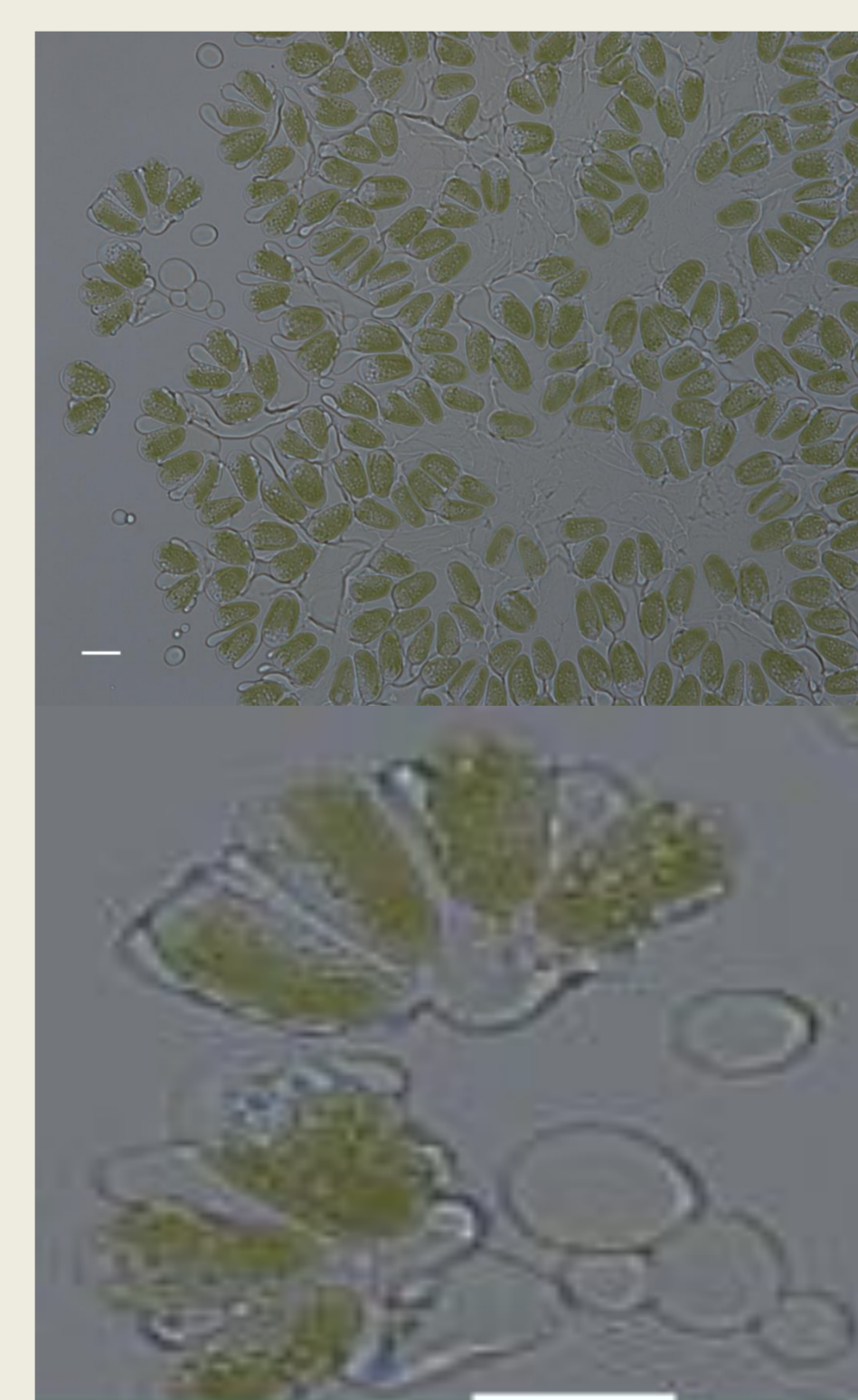
Race A SAG 30.81

Scale bar : 10µm



Race B Bot 22

Scale bar : 10µm



Conclusions - perspectives

Comparative study of the effect of shear stress on the oils composition and distribution within the extracellular polymeric matrix

- Chromatographic analysis (HPTLC)
- Morphometric analysis of the extracellular liquid hydrocarbons (Confocal microscopy)

Ishimatsu A., Matsuura, H., Sano T., Kaya K. and Watanabe M.M. 2012 Biosynthesis of isoprene units in the C34 botryococcene molecule produced by *Botryococcus braunii* strain Bot-22 Proc. Environm. Sci. 15 /56 – 65
Jin J., Duprte C., Legrand J. and Grizeau D. 2015 Characteristics of extracellular hydrocarbon-rich microalga *Botryococcus braunii* for biofuels production: advantages and constraints

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