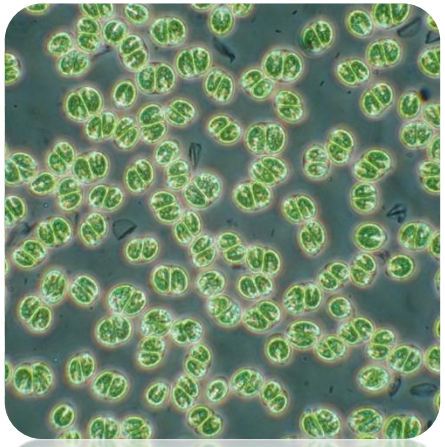
PRODUCTION OF STABLE MICROALGAE-ENRICHED FOAM

M. Vázquez, V. Si Atat, I. Garbayo, M. Cuaresma, C. Vílchez

University of Huelva




MIRACLES SPECIALTIES FROM ALGAE

This project has received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement no 613588

Universidad de Huelva

BITAL

GFT

Gesellschaft zur Förderung des Forschungs- und Technologietransfers in der Universität Bielefeld e.V.

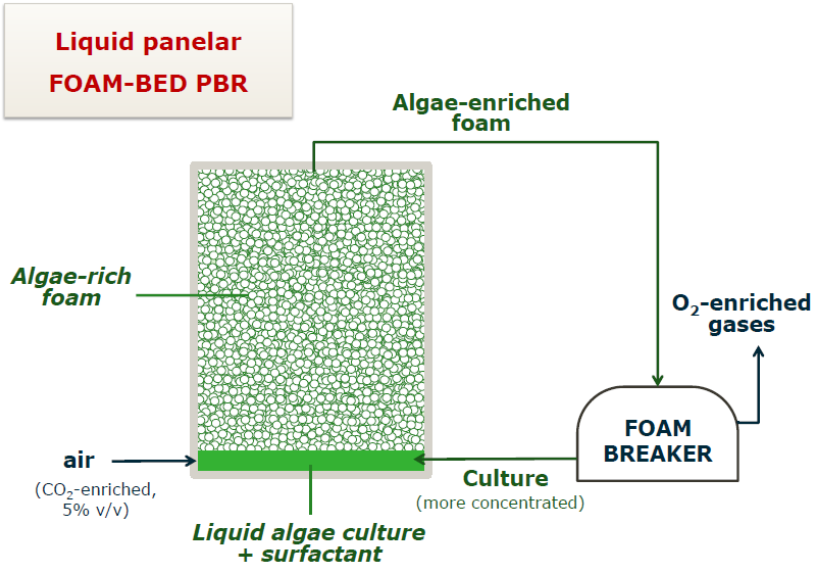
CeBiTec Center for Biotechnology

ZiF

AIM

Main objetive

- Development of a novel **Foam-Bed Photobioreactor** for cost reduction of microalgal biomass production



Specific aims *(foam formation)*

- Selection of suitable **microalgae strains**
- Determine **optimal culture conditions** to produce stable algae-rich foam

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JUSTIFICATION

Why to cultivate algae in foam?

- A foam-bed PBR is an attempt for **reducing cost of algae production** processes.

- ✓ ***High biomass productivities***
- ✓ ***Improved mass transfer***
- ✓ ***Improved light use efficiency***
- ✓ ***Dramatic reduction of water usage***
- ✓ ***Harvesting costs reduction***
- ✓ ***Avoid dewatering processes*** *like centrifugation, filtration or flocculation*

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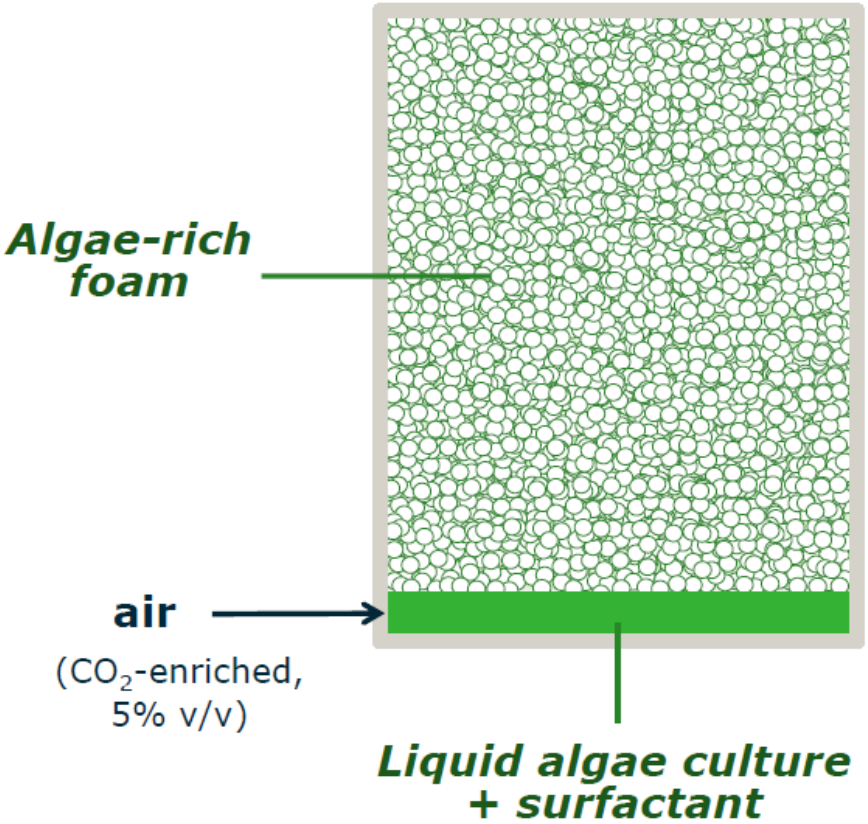
BACKGROUND

Foam flotation as an algal production system

Until now, foam flotacion has been used as a **harvesting method** (but not for cultivation!):



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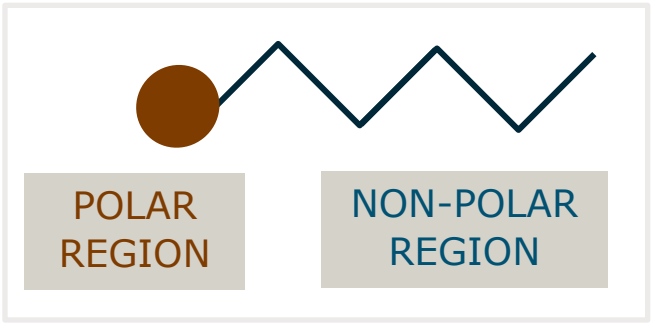

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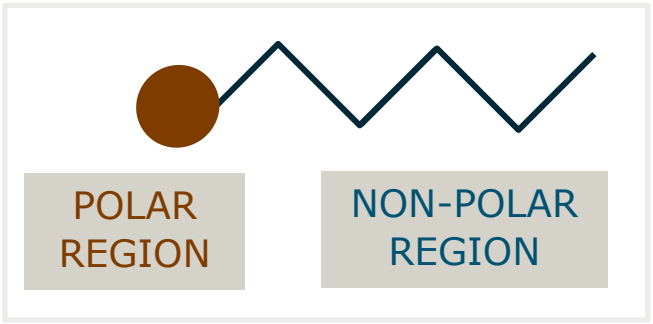

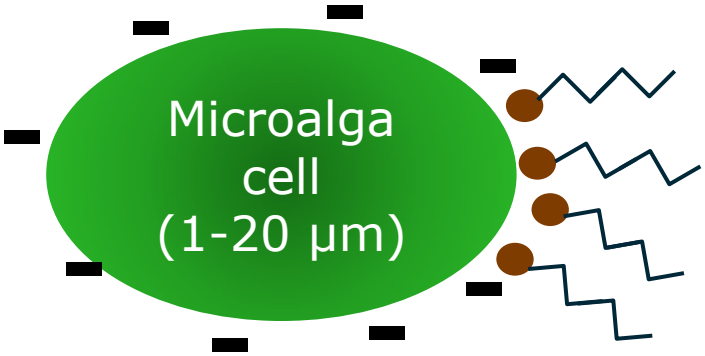

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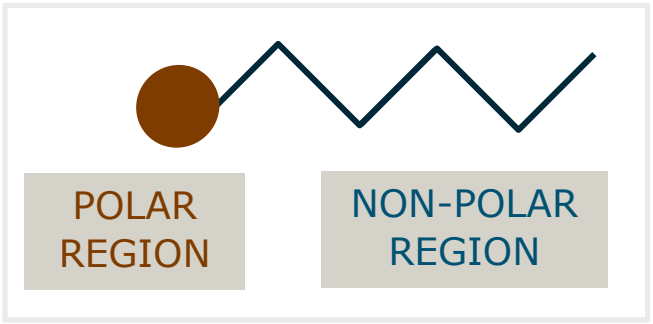

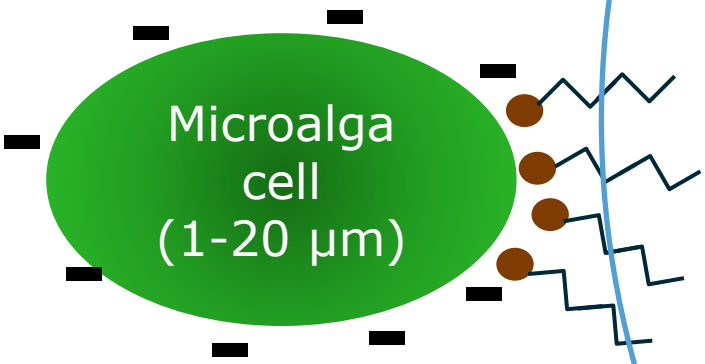

Air bubble

(700 – 1500 µm)

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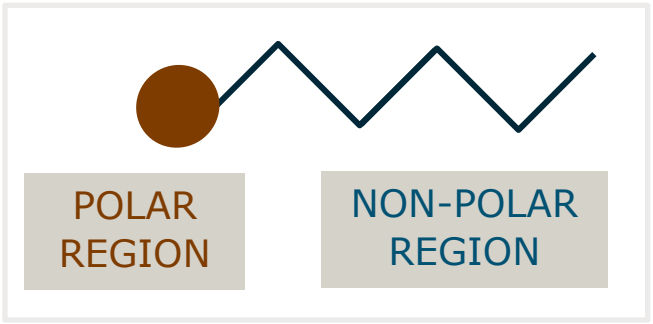

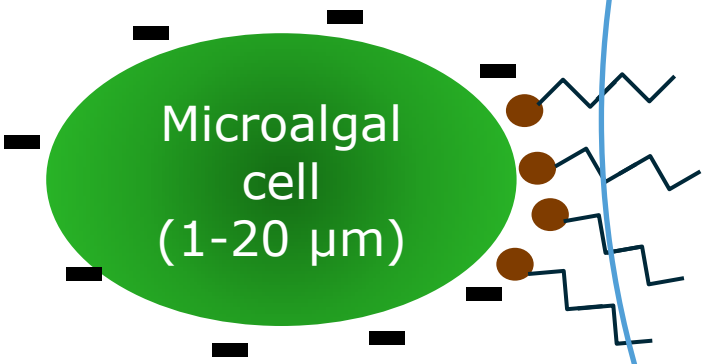

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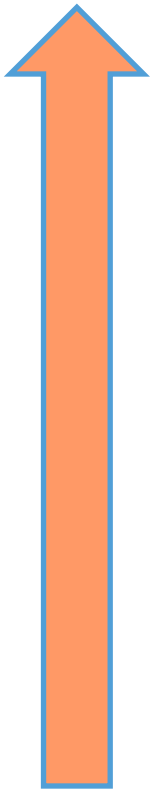

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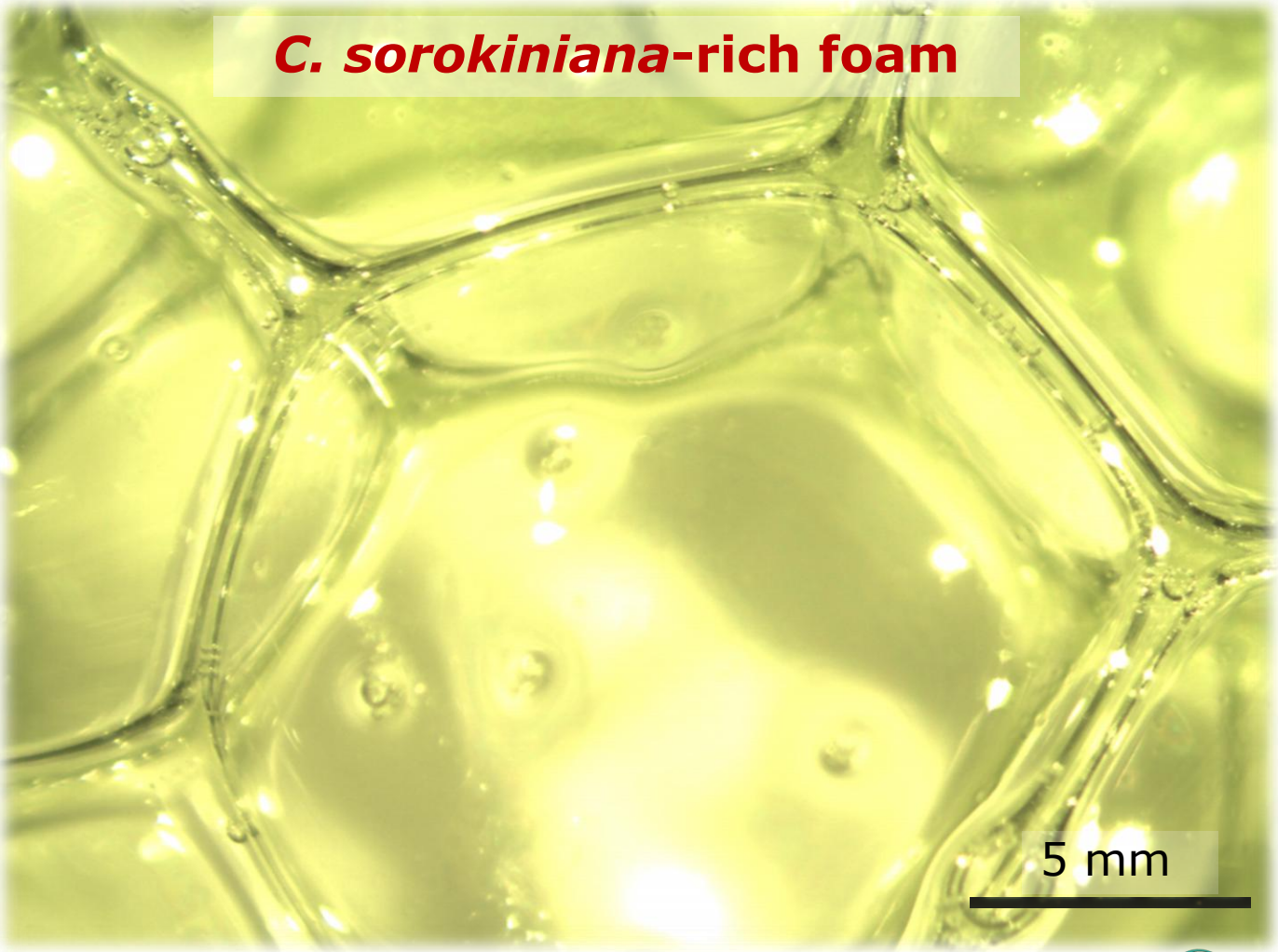

Magnifying glass 3x10

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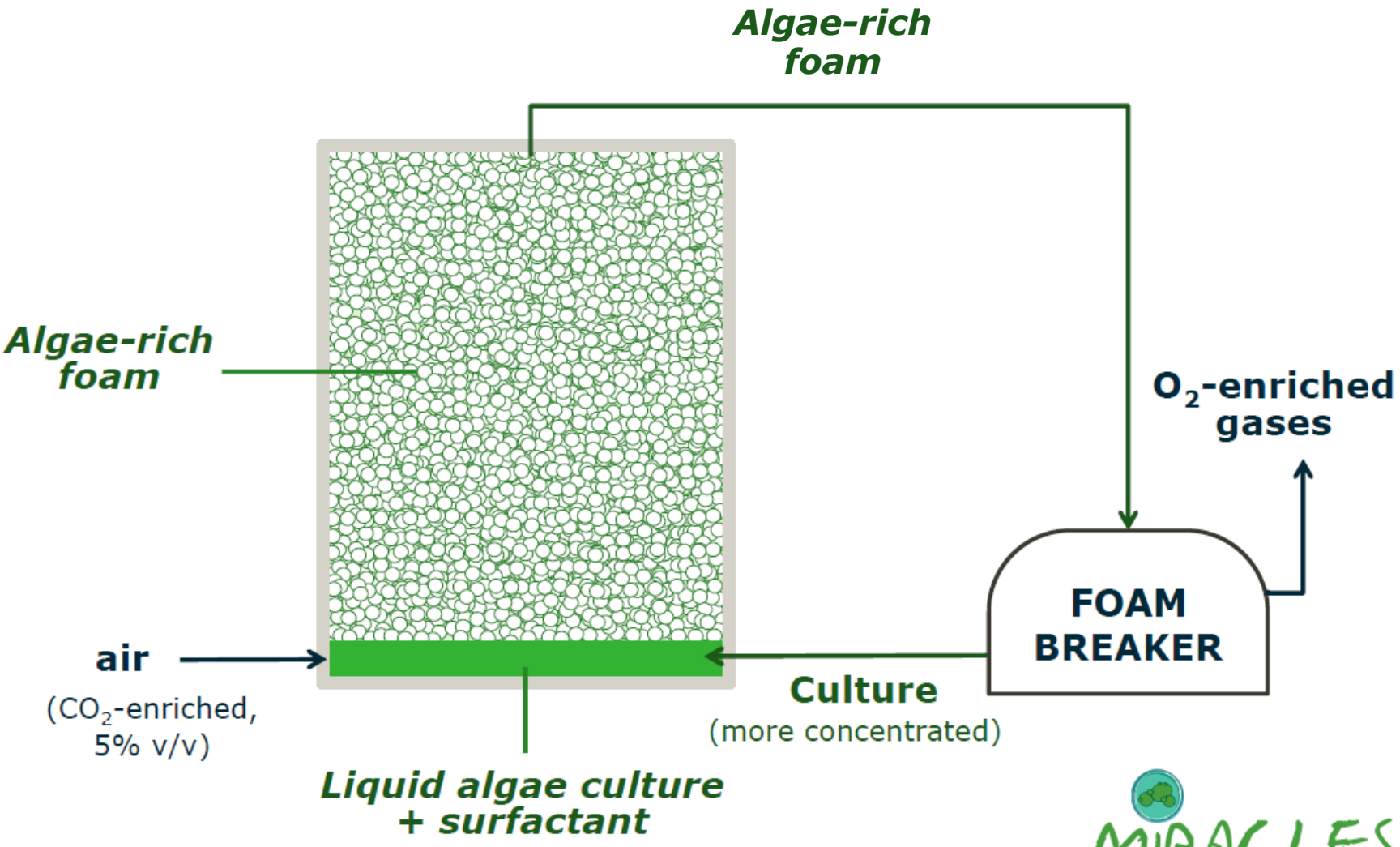

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BACKGROUND

What are the desirable conditions for producing algae-rich foam?

- ✓ Suitable choice of surfactant + microalga strain
- ✓ Microalgae hold-up ⟶ Algae-enriched foam
- ✓ Microalgae viability
- ✓ Stable foam
- ✓ Net foam accumulation

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Key issues

- ✓ Reactor design and operational conditions
- ✓ Surfactant
- ✓ Microalgae culture

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What are the factors of the microalgae culture which influence the foam formation process?

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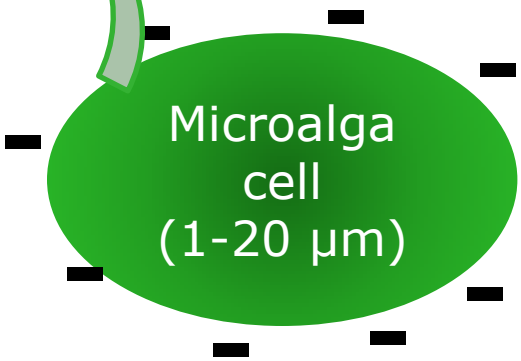
ALGAL CELL



- Algal cell size
- Cell surface hydrophobicity
- Culture viscosity (biomass concentration)
- Culture age
- Production of EPS

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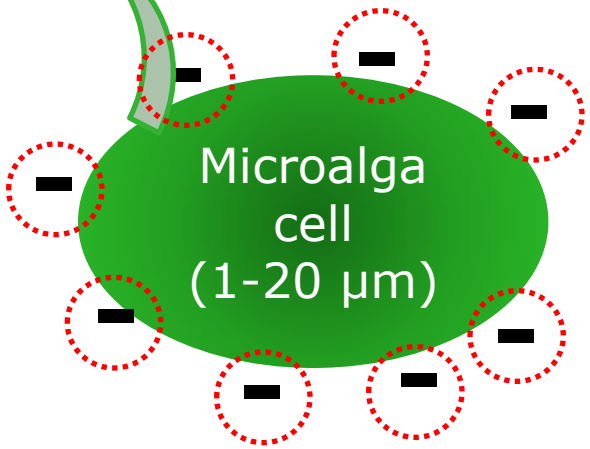

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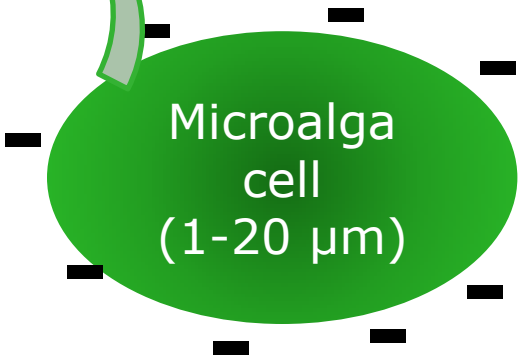

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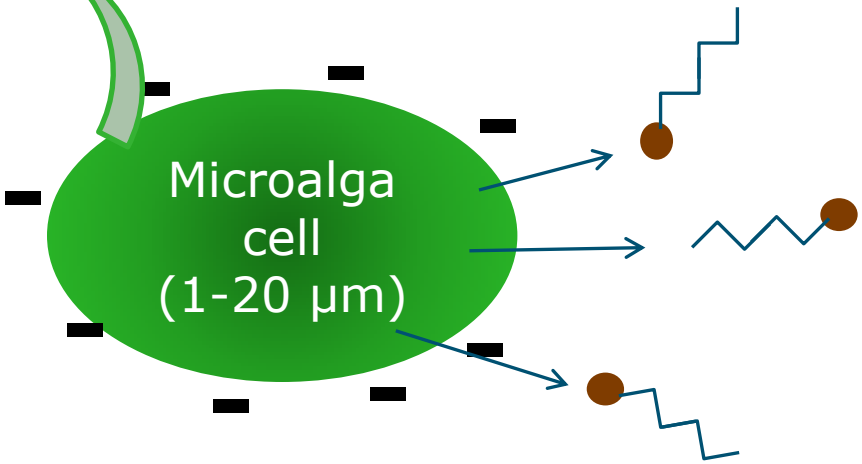

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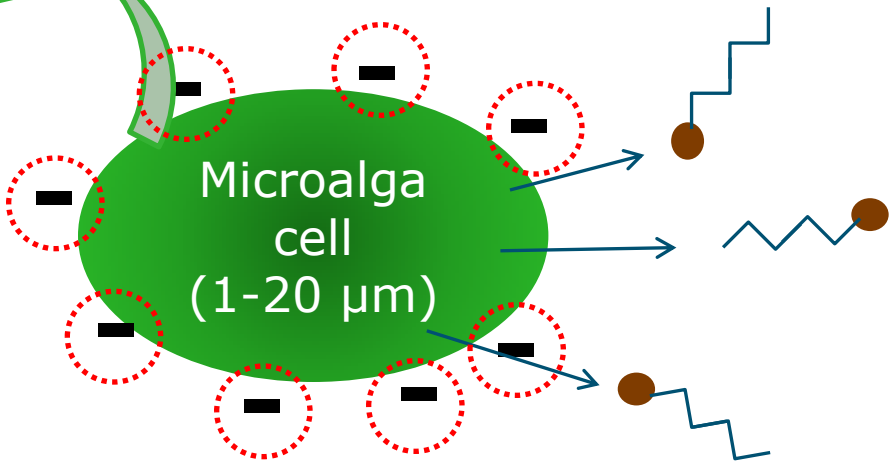

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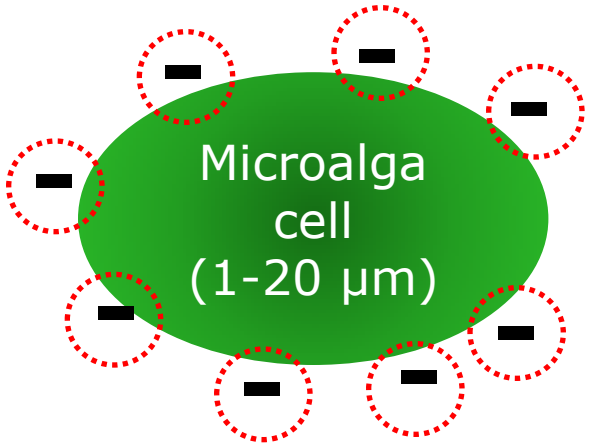

CULTURE MEDIUM

- **pH**
- Culture Medium salinity

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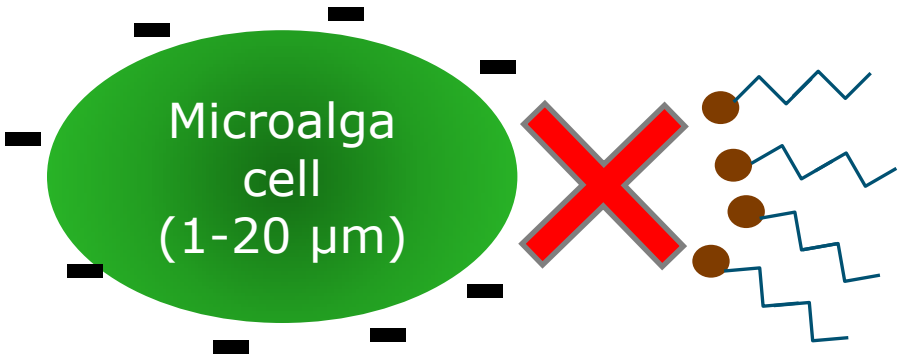

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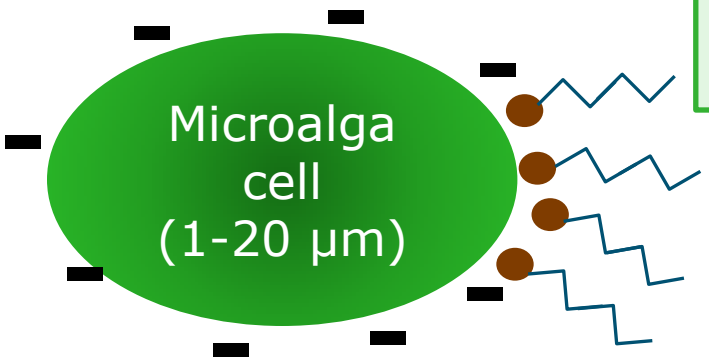

SURFACTANT INTERACTION

CULTURE MEDIUM

- pH
- Culture Medium salinity

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RESULTS

What have we done?

1. Preselection and production of **microalgae species** suitable for natural foam production.
2. Assessment of **natural foam production**
3. Comparison of surfactant-mediated **foam production speeds** (up-flow)
4. Study of the **hold-up ability**
5. Assessment of **foam stability**.

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MIRACLES
SPECIALTIES FROM ALGAE

RESULTS

1. Preselection and production of microalgae species suitable for natural foam production.

- 8 microalgae species
- Literature survey
- Strains included in the project
- Own experience and foaming observations

Microalga	Culture medium	Extracelullar polymers	References
Nannochloropsis gaditana (CCFM)	F/2	Polysaccharides	Guerrero et al. 2004
Scenedesmus obliquus (CCFM)	BG-11	Proteins, polysaccharides	Corradi et al. 1998
Phaeodactylum tricornutum (CCFM)	F/2	Proteins, polysaccharides	Willis et al. 2013; Bruckner et al. 2011
Isochrysis galbana (CCFM)	F/2	-	-
Chlorella sorokiniana	M-8a	Proteins, polysaccharides	Wang et al. 2004; Campos et al. 2013
Neochloris oleoabundans	F/2	Biopolymers	Wu et al. 2011
Botryococcus braunii • ***var*. Showa** • **(B14)**	Chu	Hydrocarbons, exopolysaccharides	Dayananda et al. 2007

RESULTS

1. Preselection and production of microalgae species suitable for natural foam production.

Stable repeated-batch cultures

Culture conditions

Light intensity:	160 μmol·m^{-2}·s^{-1}
Temperature:	26 celsius
Mode:	Repeated-batch, log phase
Bubbling:	CO_2 in air (5% v/v)
Volume:	2 to 5 L
Dense cultures:	Centrifugation

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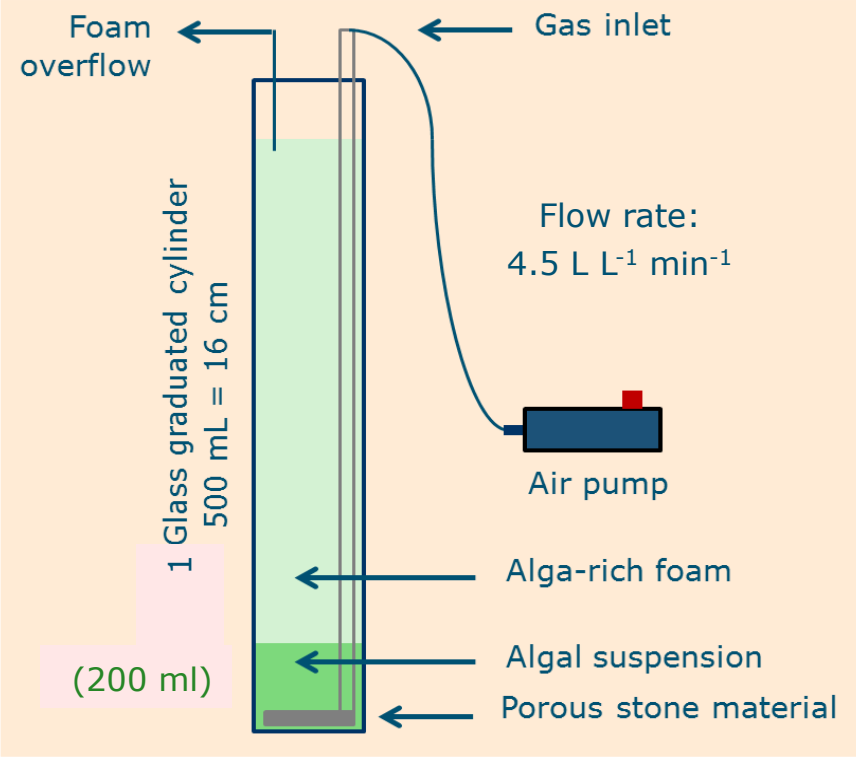
2. Assessment of natural foam production

Comparing ability to produce foam naturally and in a fast way



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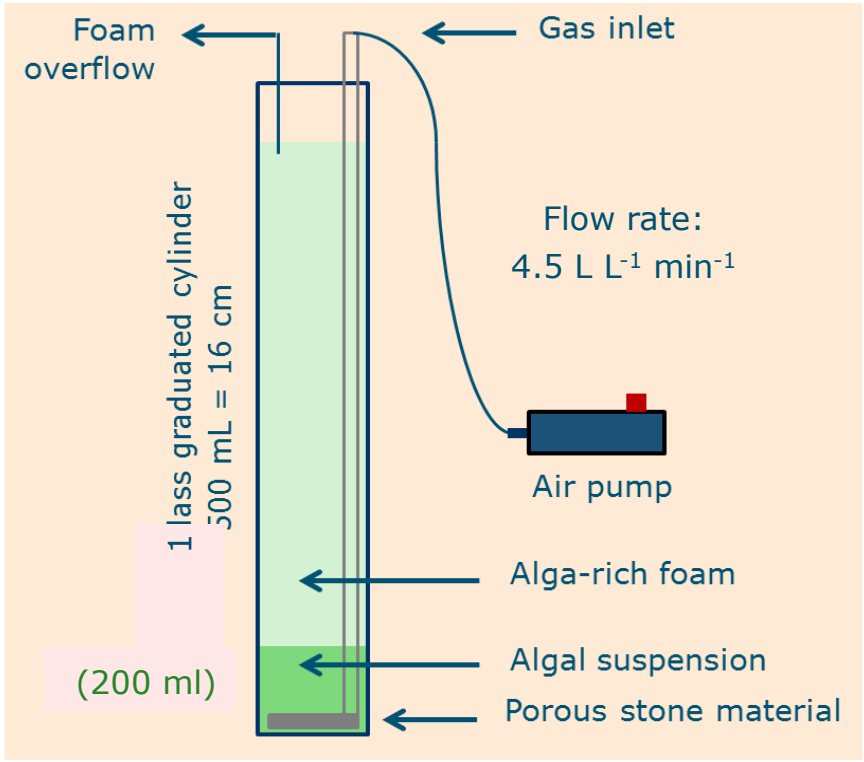

Study parameters:

- Microalgae strains

- Freshwater
  - *C. sorokiniana*
  - *S. obliquus,*
  - *B. braunii var.* Showa
  - *B. braunii* (B14)
- Marine
  - *I. galbana*
  - *N. gaditana*
  - *P. tricornutum*
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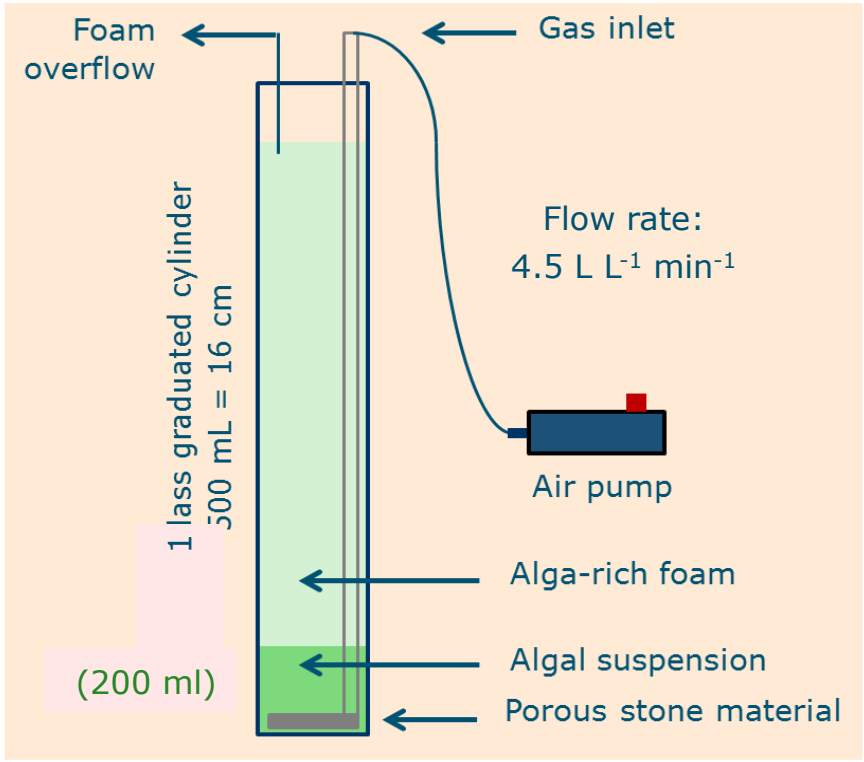

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- Growth phase

- Exponential
- Stationary

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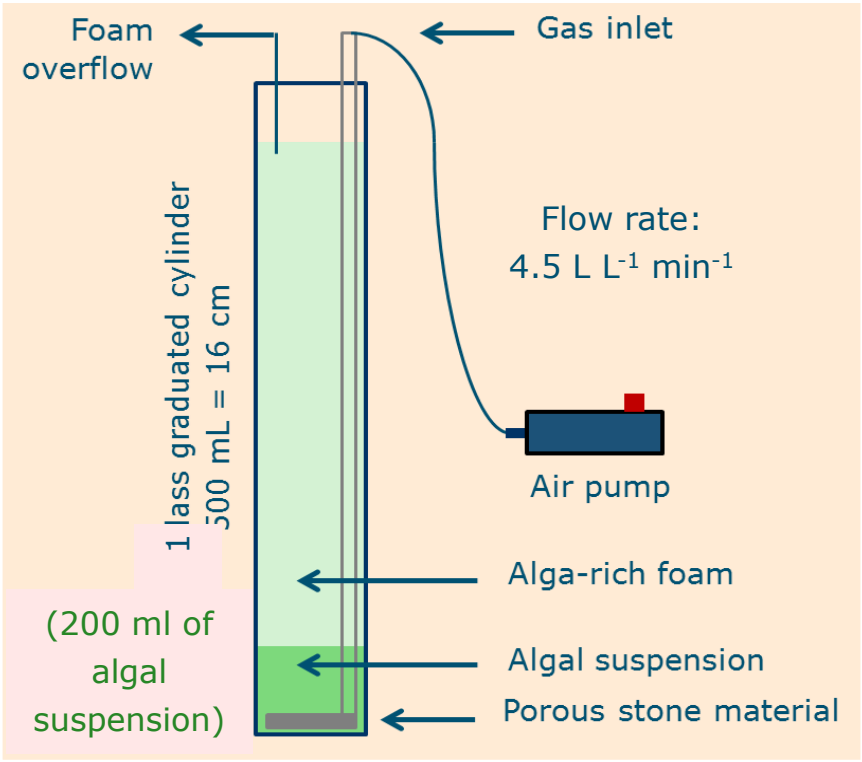

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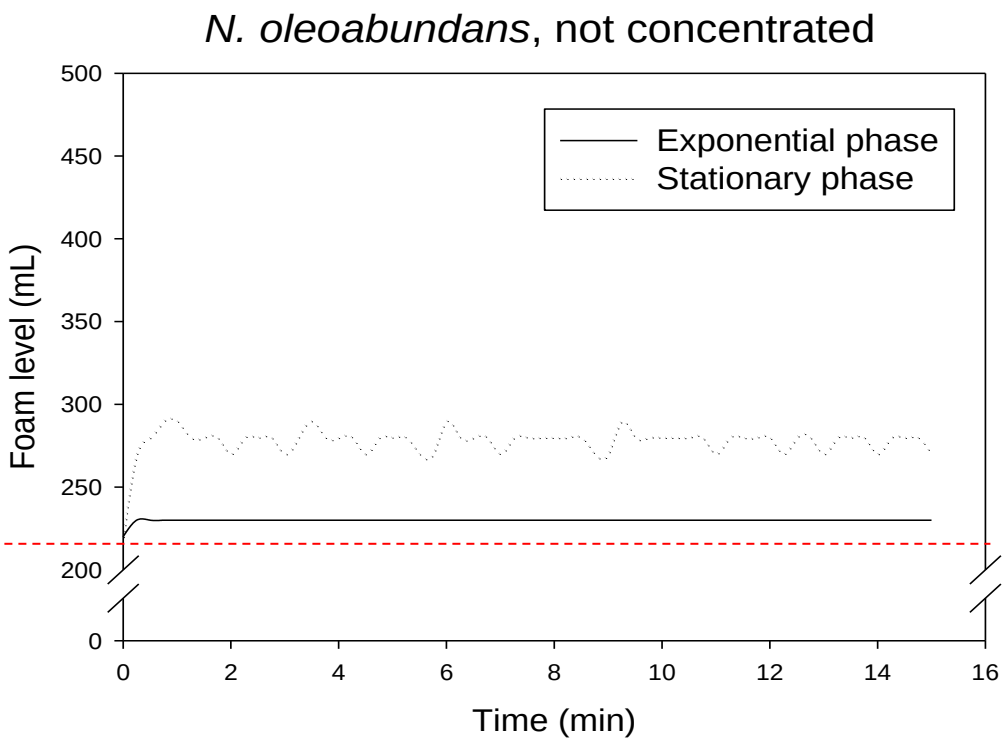
- Biomass concentration

- Normal
- 3-time concentrated

RESULTS

Effect of growth phase on natural foaming

Initial level= 210 ml (200ml of algal suspension + diffussers volume)



✓ **Stationary > Exponential**: *N. oleoabundans, B. braunii* var. Showa*, N. gaditana*

✓ **Foaming is preferably in exponential phase**

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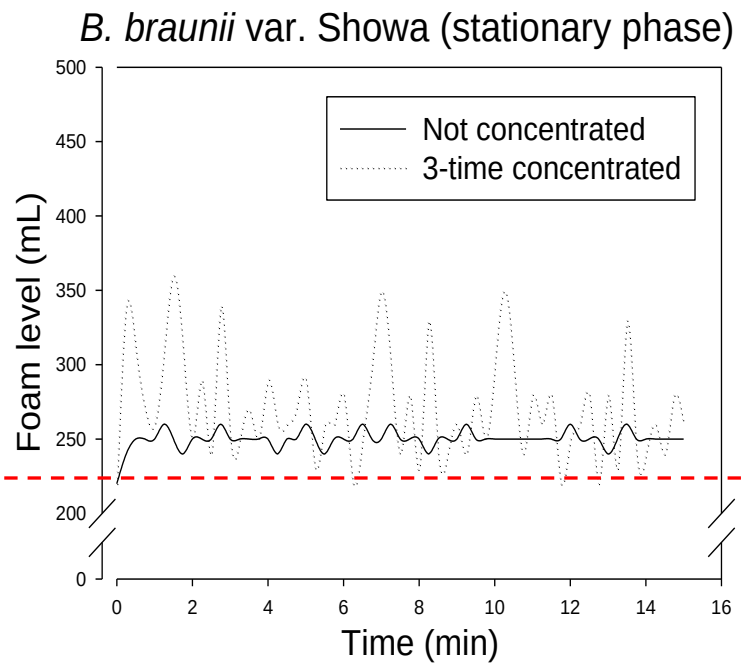

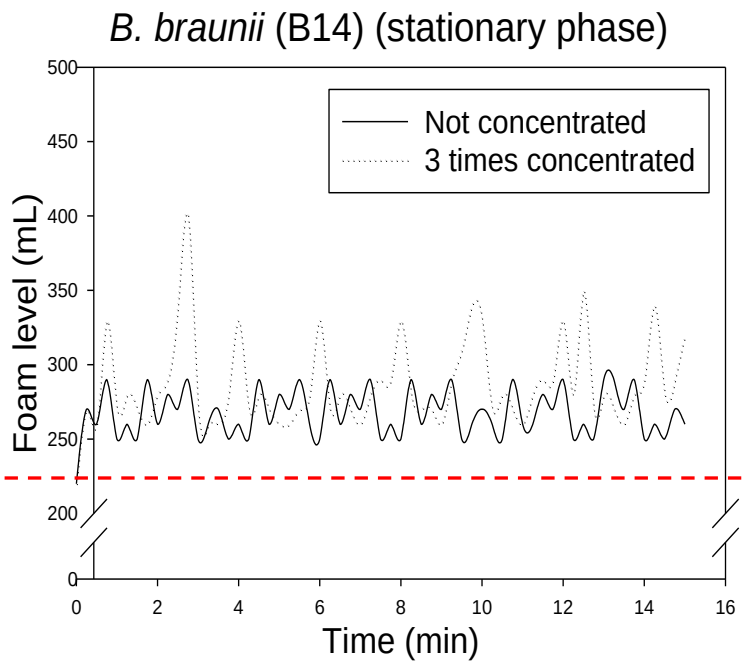

✓ **Positive effect**: *B. braunii* var. Showa and *B. braunii* (B14)

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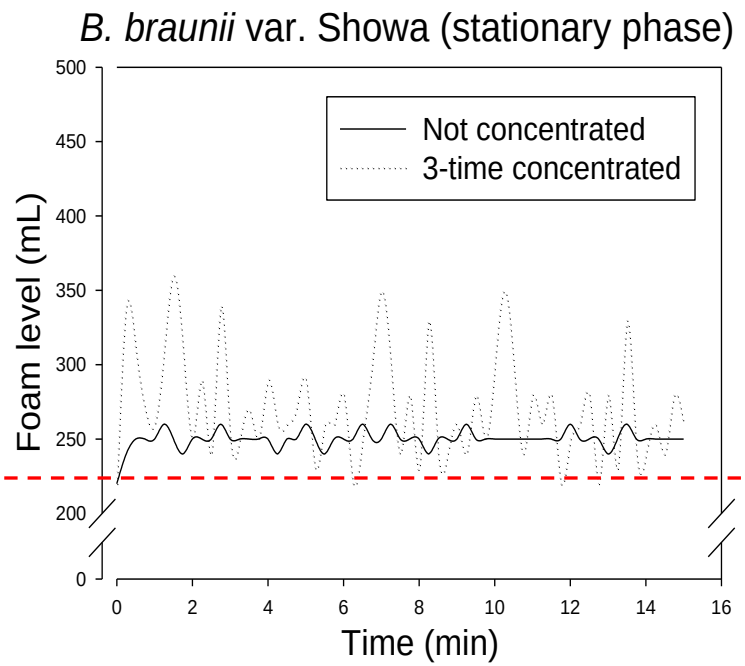

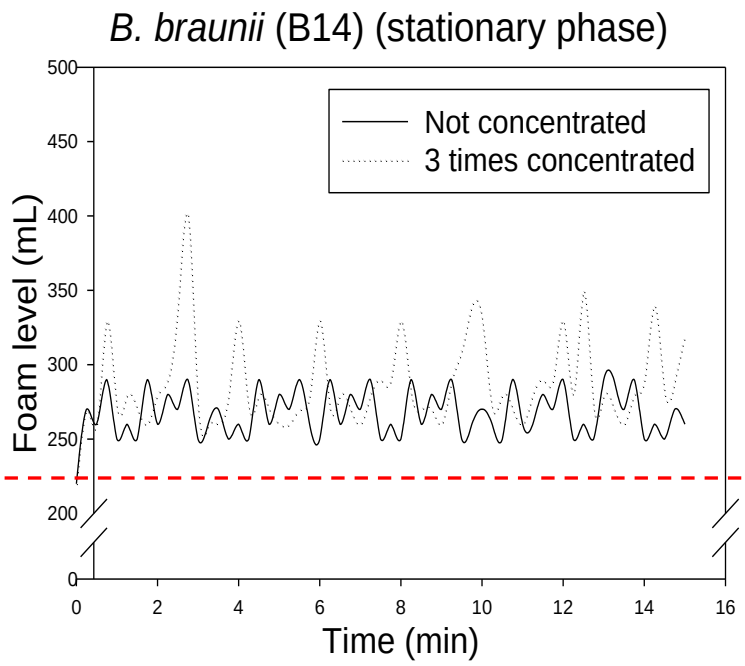

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Not stable foam naturally produced in any case.

SURFACTANT ADDITION IS REQUIRED !!!

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3. Comparison of **surfactant-mediated foam production speeds (up-flow)**
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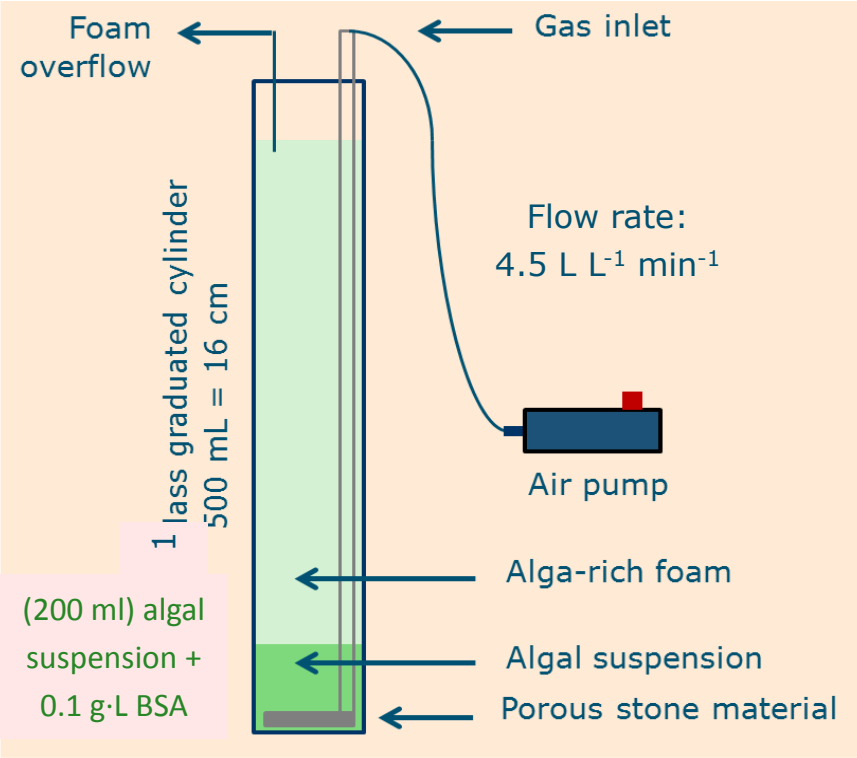
MIRACLES
SPECIALTIES FROM ALGAE

3. Surfactant-mediated foam production speeds (up-flow)

Comparing ability to produce BSA-mediated foaming in a fast way



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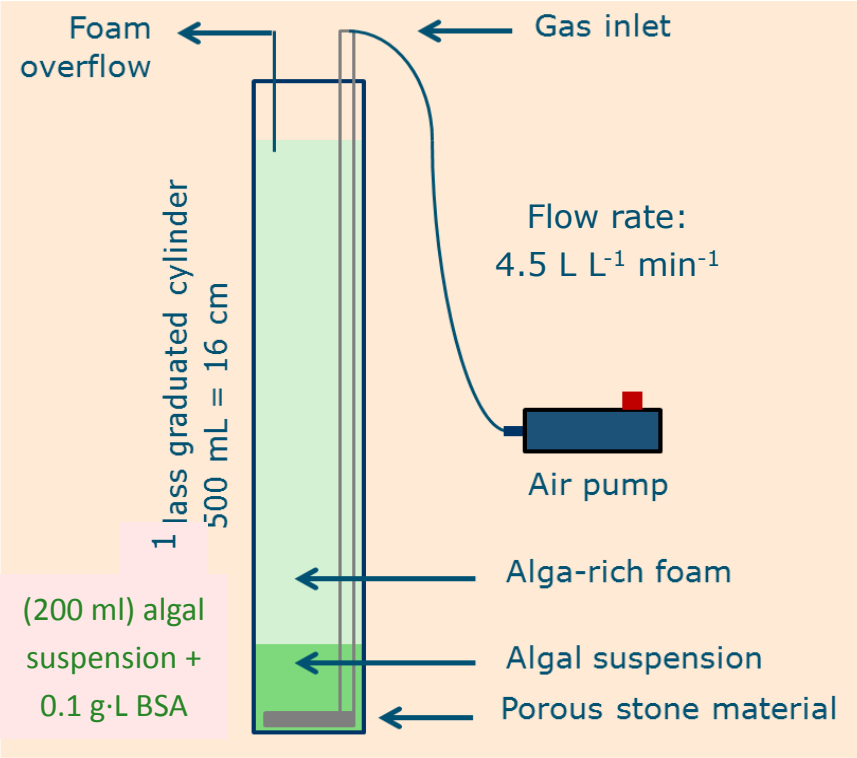

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BSA: Bovine Serum Albumin

- ✓ Organic
- ✓ Non-toxic
- ✓ Efficacy as surfactant previously shown

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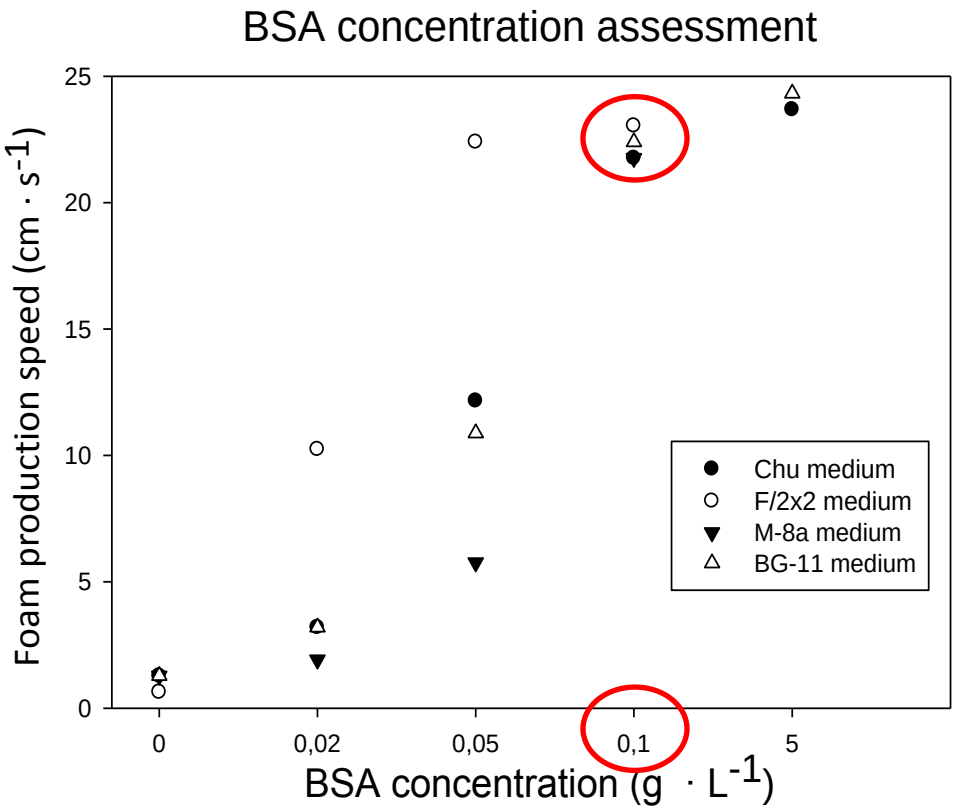

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- ✓ **0.1 g · L^{-1}**

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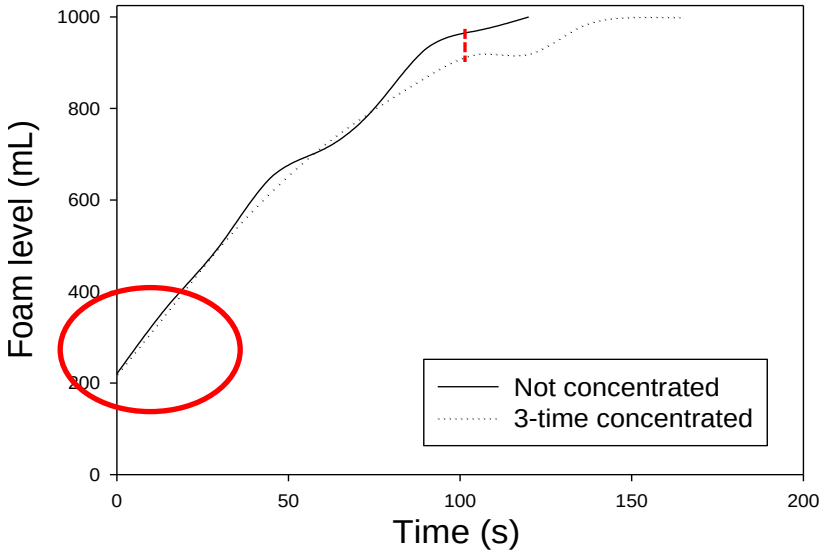
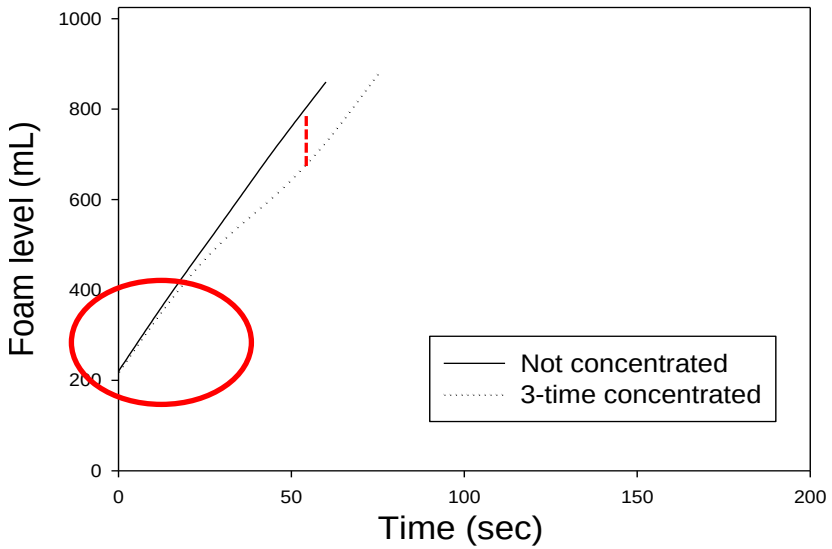
Effect of biomass concentration on BSA-mediated foam up-flow

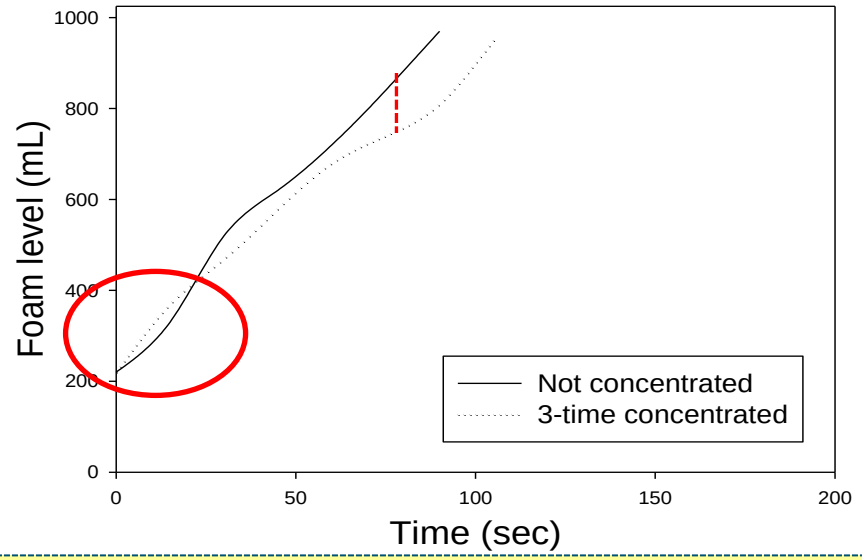
B. braunii (B14), stationary phase



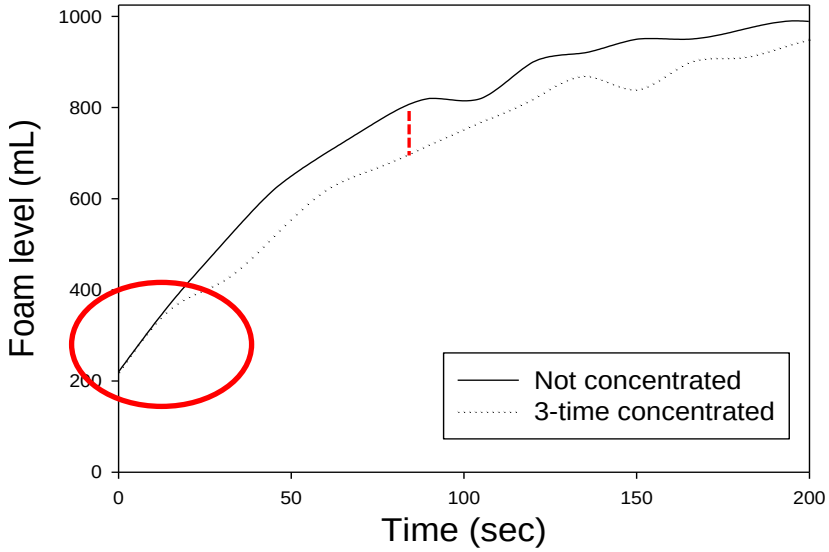
C.sorokiniana, exponential phase



N. oleoabundans, stationary phase



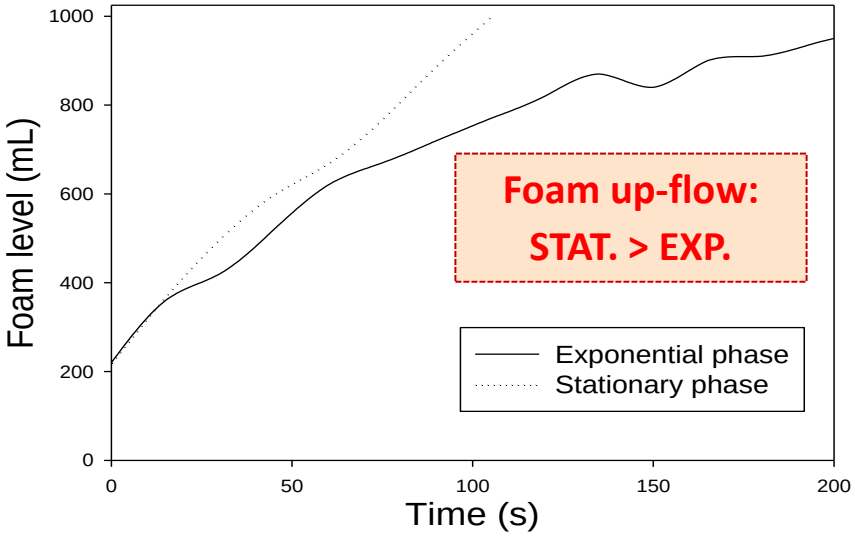
N. gaditana, exponential phase



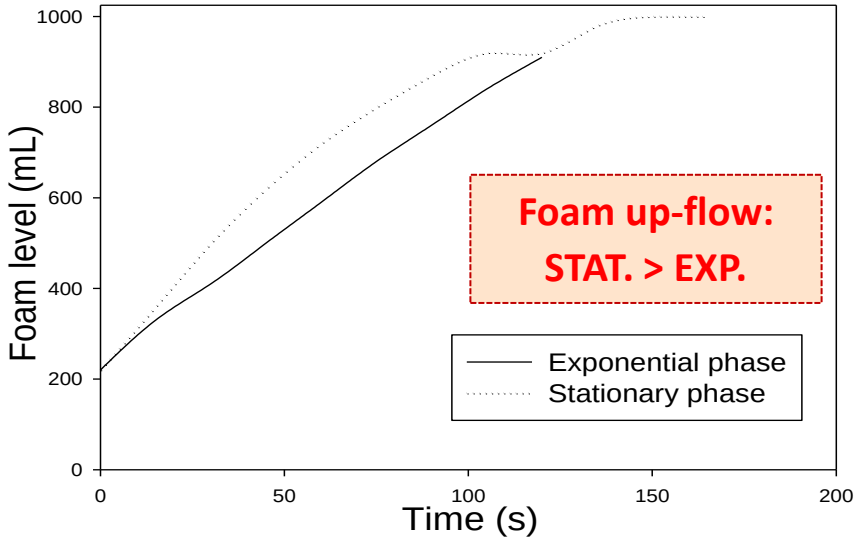
Biomass preconcentration led to lower up-flow just seconds after foaming started up

Effect of growth phase on BSA-mediated foam up-flow

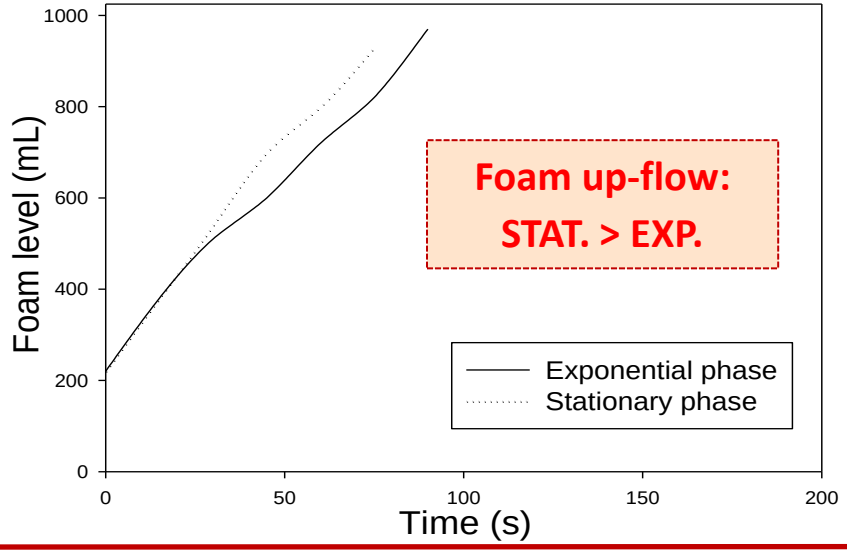
N. gaditana, 3-time concentrated



B. Braunii (B14), 3-time concentrated



S. obliquus, 3-time concentrated

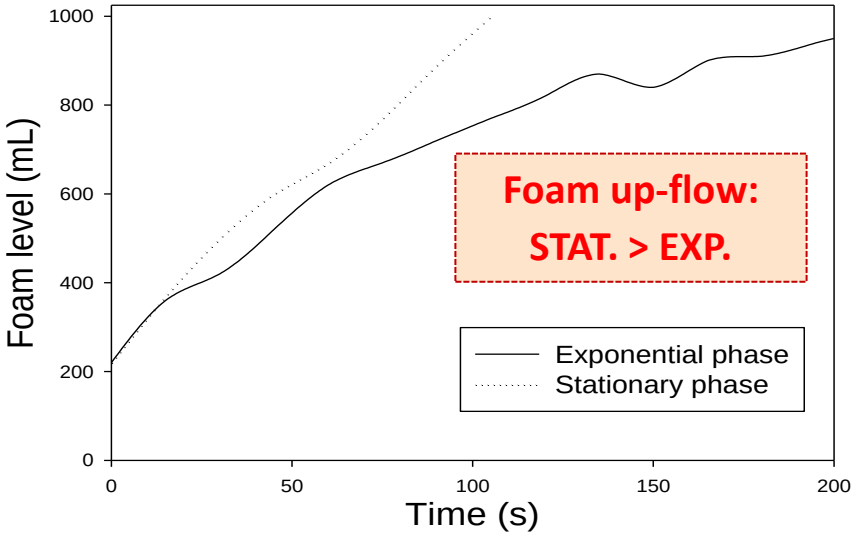


Stationary phase favoured foaming in most cases

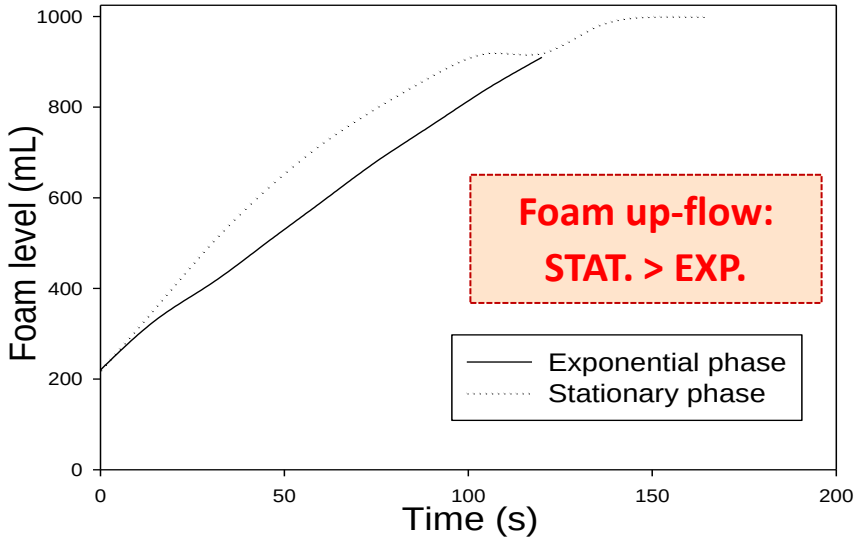
MIRACLES
SPECIALTIES FROM ALGAE

Effect of growth phase on BSA-mediated foam up-flow

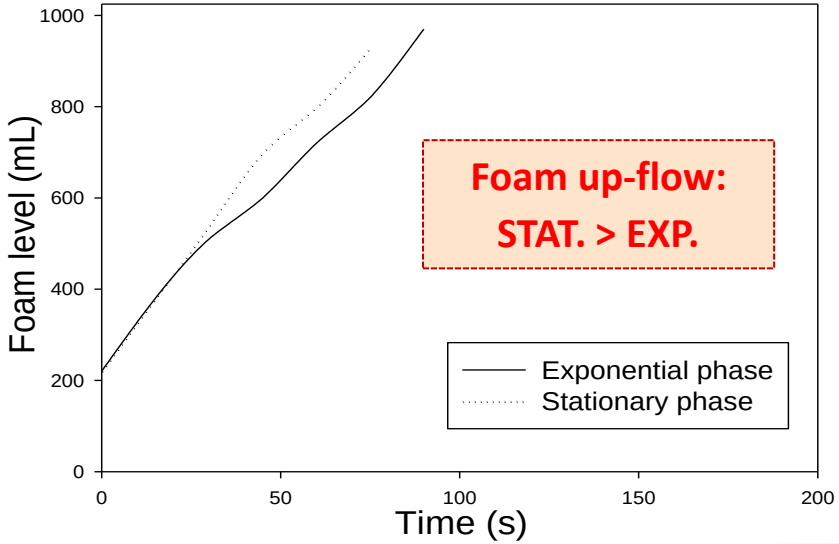
N. gaditana, 3-time concentrated



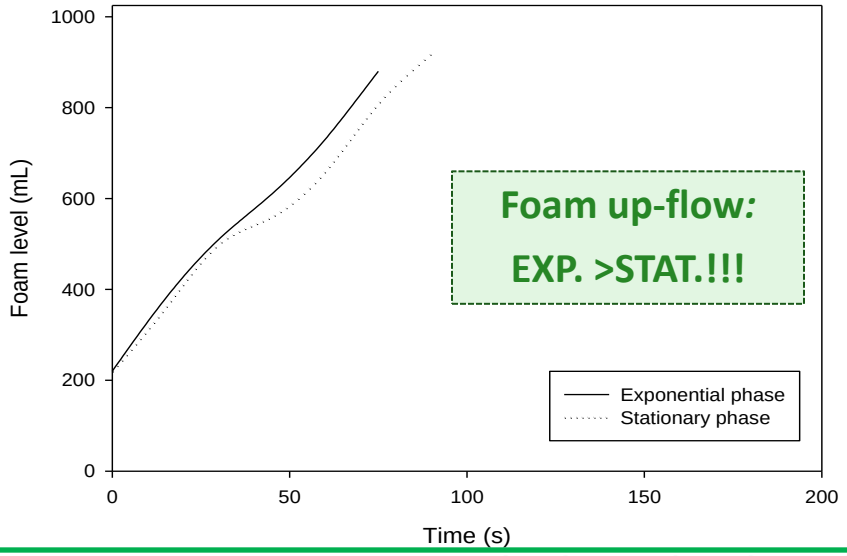
B. Braunii (B14), 3-time concentrated



S. obliquus, 3-time concentrated

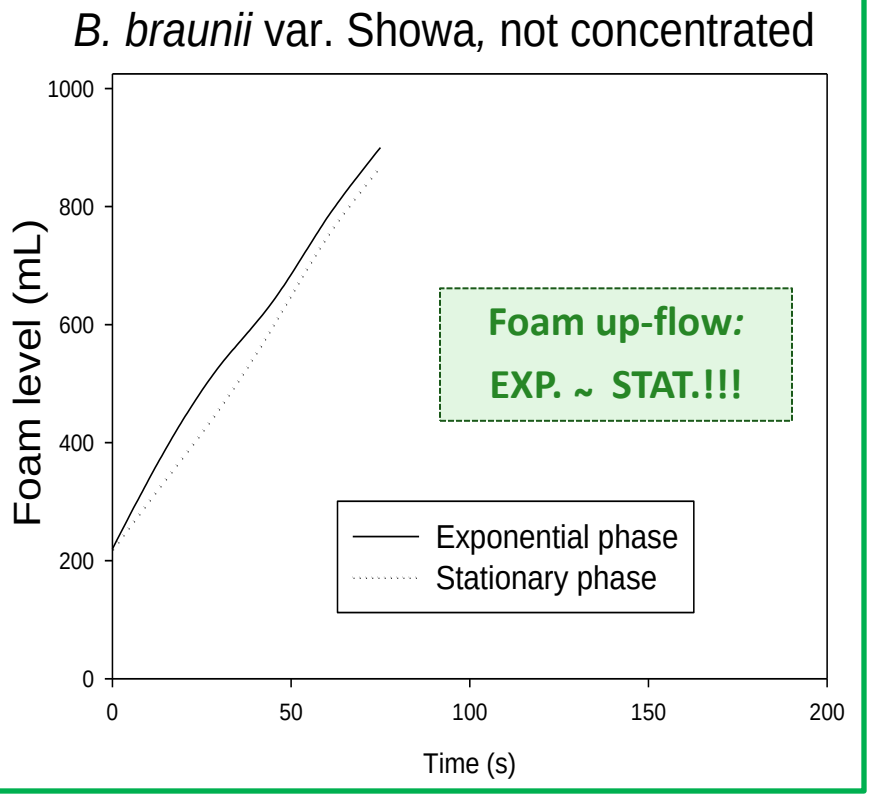


C. sorokiniana, 3-time concentrated



SPECIALTIES FROM ALGAE

B. braunii var. Showa produces foam naturally, even in exponential phase…



However… _Showa_ strain will not be considered as a suitable strain to produce algae-rich foam because of its low hold-up.

MIRACLES
SPECIALTIES FROM ALGAE

BSA-mediated foam up-flow speeds (cm · s^{-1})

	Chu medium	F/2x2 medium	*Chlorella sorokiniana*		*Botryococcus braunii* var. Showa	
	NO ALGAE	NO ALGAE	LOW ALGAE CONC.	HIGH ALGAE CONC.	LOW ALGAE CONC.	HIGH ALGAE CONC.
Exponential	0.36	0.38	**0,35**	0.31	**0,33**	0,32
Stationary			0.32	0.30	0.26	0.26
	Botryococcus braunii (B14)		*Scenedesmus obliquus*		*Nannochloropsis gaditana*	
	LOW ALGAE CONC.	HIGH ALGAE CONC.	LOW ALGAE CONC.	HIGH ALGAE CONC.	LOW ALGAE CONC.	HIGH ALGAE CONC.
Exponential	0,21	0,20	**0,31**	0,31	**0,30**	0,21
Stationary	0,30	0,30	0,33	0,34	0,30	0,30
	Isochrysis galbana		*Neochloris oleoabundans*		*Phaeodactylum tricornutum*	
	LOW ALGAE CONC.	HIGH ALGAE CONC.	LOW ALGAE CONC.	HIGH ALGAE CONC.	LOW ALGAE CONC.	HIGH ALGAE CONC.
Exponential	N.M.	N.M.	0,23	0,23	0,27	0,32
Stationary	0,18	0,13	0,32	0,27	N.M.	N.M.

MIRACLES
SPECIALTIES FROM ALGAE

RESULTS

What have we done?

1. Preselection and production of **microalgae species** suitable for natural foam production.
2. Assessment of natural foam production
3. Comparison of surfactant-mediated foam production speeds (up-flow)
4. Study of **hold-up ability**
5. Assessment of **foam stability**.

MIRACLES
SPECIALTIES FROM ALGAE

4. Study of algae hold-up abilities

Study parameters:

- Microalgae strains

- Freshwater
  - *C. Sorokiniana*
  - *S. Obliquus,*
  - *B. braunii var.* Showa
  - *B. braunii* (B14)
- Marine
  - *I. galbana*
  - *N. gaditana*
  - *P. tricornutum*
  - *N.oleoabundans*

MIRACLES
SPECIALTIES FROM ALGAE

4. Study of algae hold-up abilities

Study parameters:

- Microalgae strains

- Freshwater
  - *C. Sorokiniana*
  - *S. Obliquus,*
  - *B. braunii var.* Showa
  - *B. braunii* (B14)
- Marine
  - *I. galbana*
  - *N. gaditana*
  - *P. tricornutum*
  - *N.oleoabundans*

- Growth phase

- Exponential
- ~~Stationary~~

MIRACLES
SPECIALTIES FROM ALGAE

4. Study of algae hold-up abilities

Study parameters:

- Microalgae strains

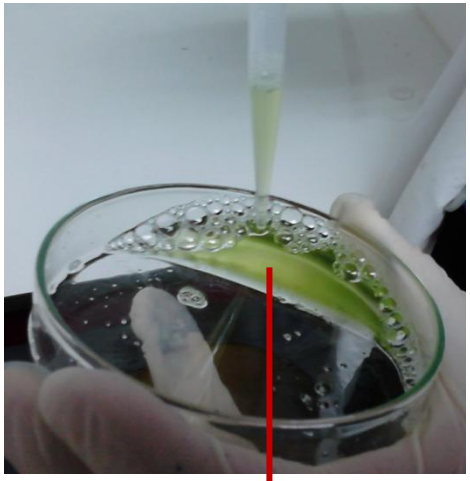
- Freshwater
  - *C. Sorokiniana*
  - *S. Obliquus,*
  - *B. braunii var.* Showa
  - *B. braunii* (B14)
- Marine
  - *I. galbana*
  - *N. gaditana*
  - *P. tricornutum*
  - *N.oleoabundans*

- **Growth phase**
  - Exponential
  - Stationary
- **Biomass concentration**
  - *LOW BIOMASS: 1.5-2 g · L^{-1}*
  - *HIGH BIOMASS: 8 – 10 g · L^{-1}*

MIRACLES
SPECIALTIES FROM ALGAE

4. Study of algae hold-up abilities

Comparing hold-up abilities



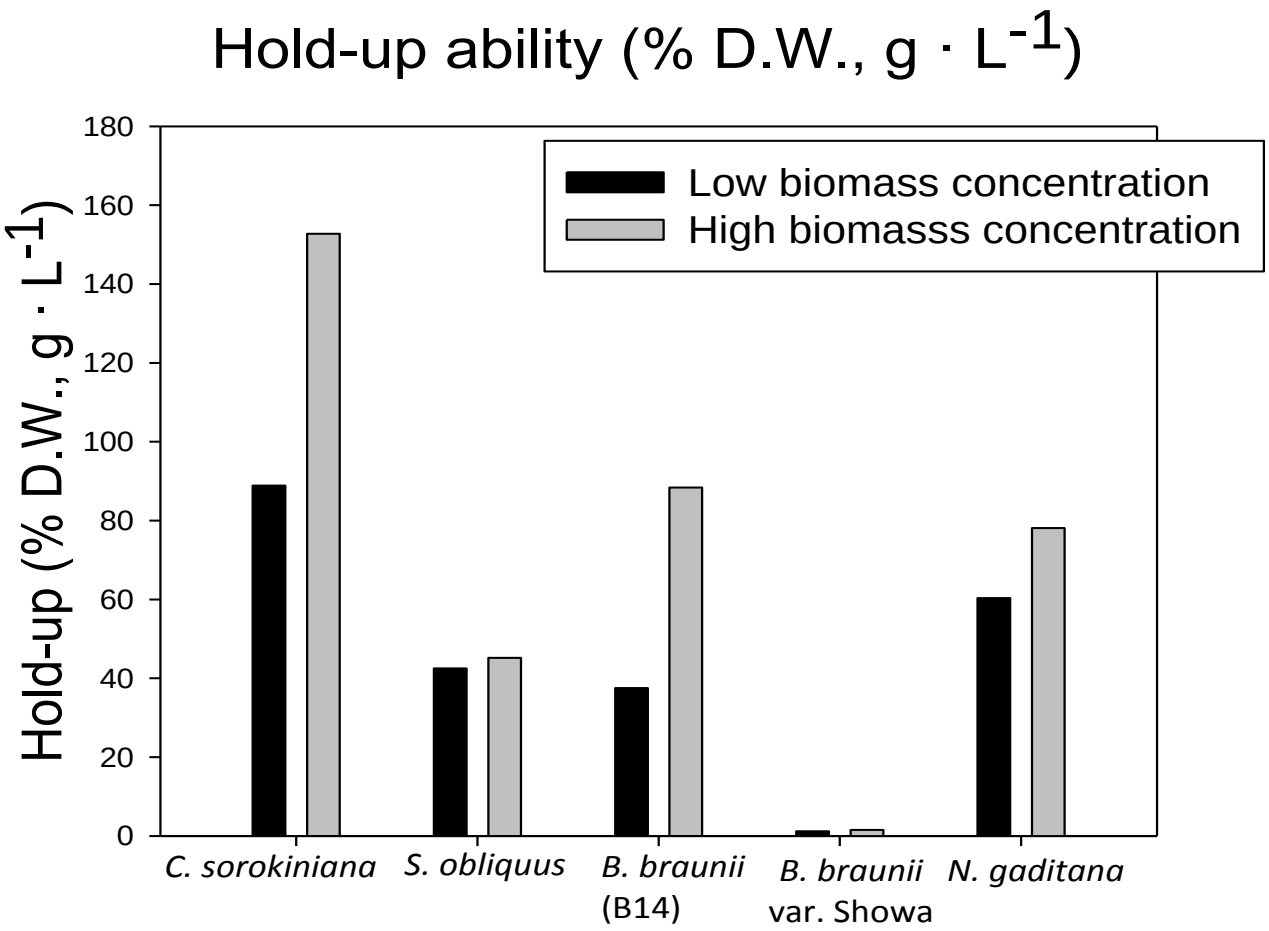
$D.W.\ (g \cdot L^{-1})_{liquid\ suspension}$

$D.W.\ (g \cdot L^{-1})_{liquid\ from\ the\ foam}$

$$\text{Hold-up (\%)} = \frac{D.W.\ (g \cdot L^{-1})_{liquid\ from\ the\ foam}}{D.W.\ (g \cdot L^{-1})_{liquid\ suspension}} \times 100$$

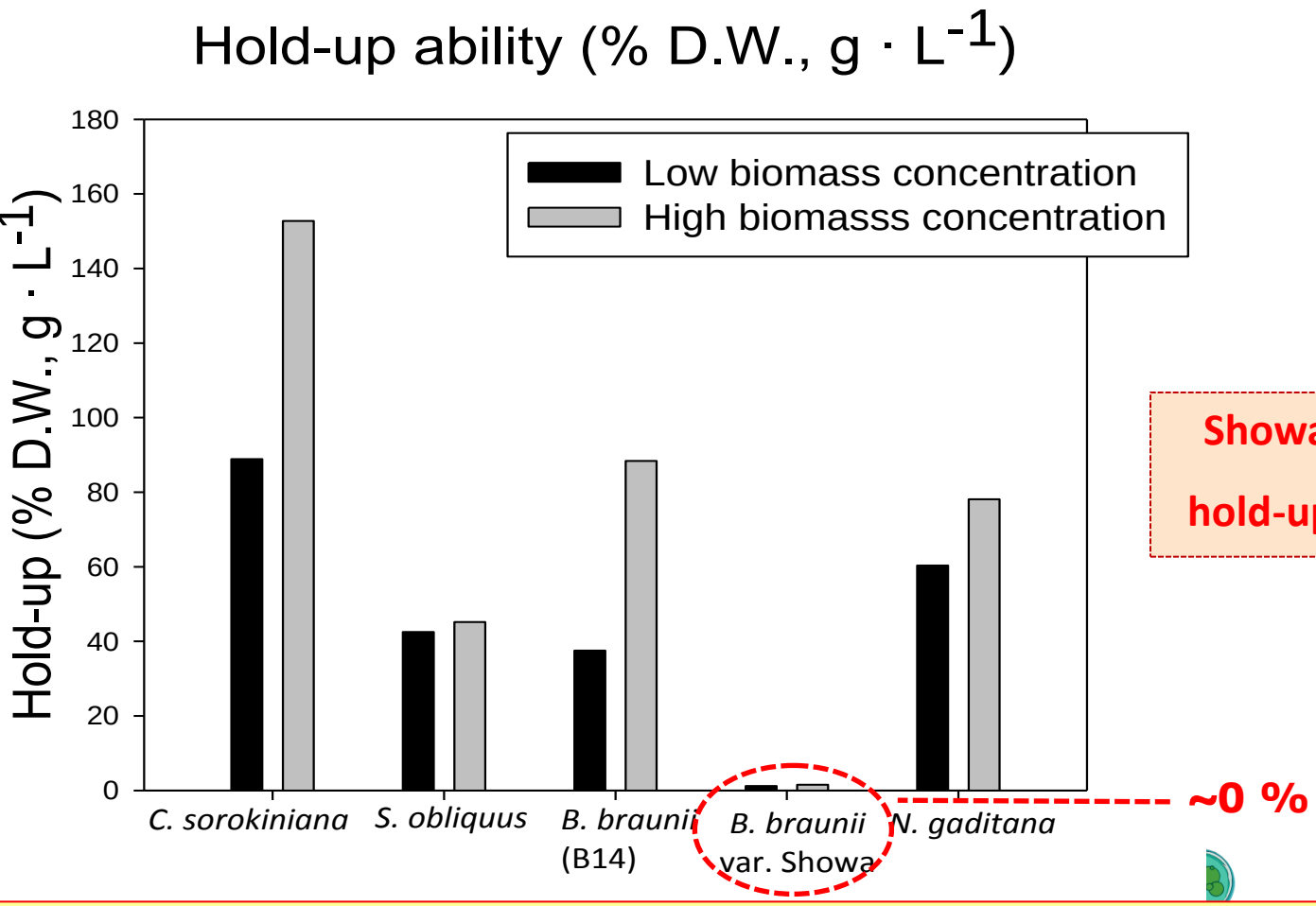
MIRACLES
SPECIALTIES FROM ALGAE

4. Study of algae hold-up abilities



Biomass preconcentration always favoured algae hold-up abilities.

4. Study of algae hold-up abilities



Showa strain

hold-up ~ 0%.

Biomass preconcentration always favoured algae hold-up abilities.

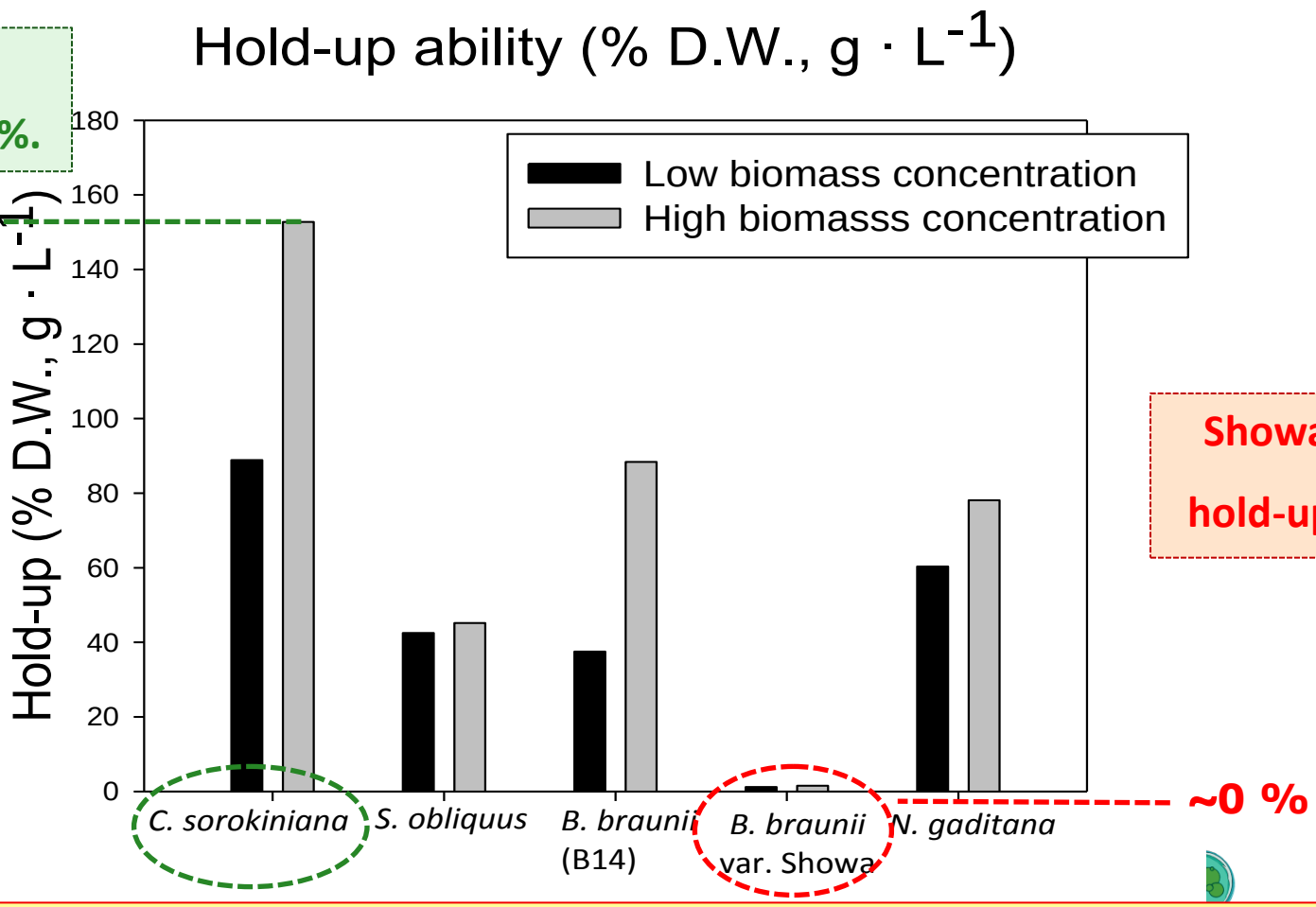
RESULTS

4. Study of algae hold-up abilities

Chlorella

hold-up>100%.

~150 %



~0 %

Showa strain

hold-up ~ 0%.

Biomass preconcentration always favoured algae hold-up abilities.

SPECIALTIES FROM ALGAE

RESULTS

What have we done?

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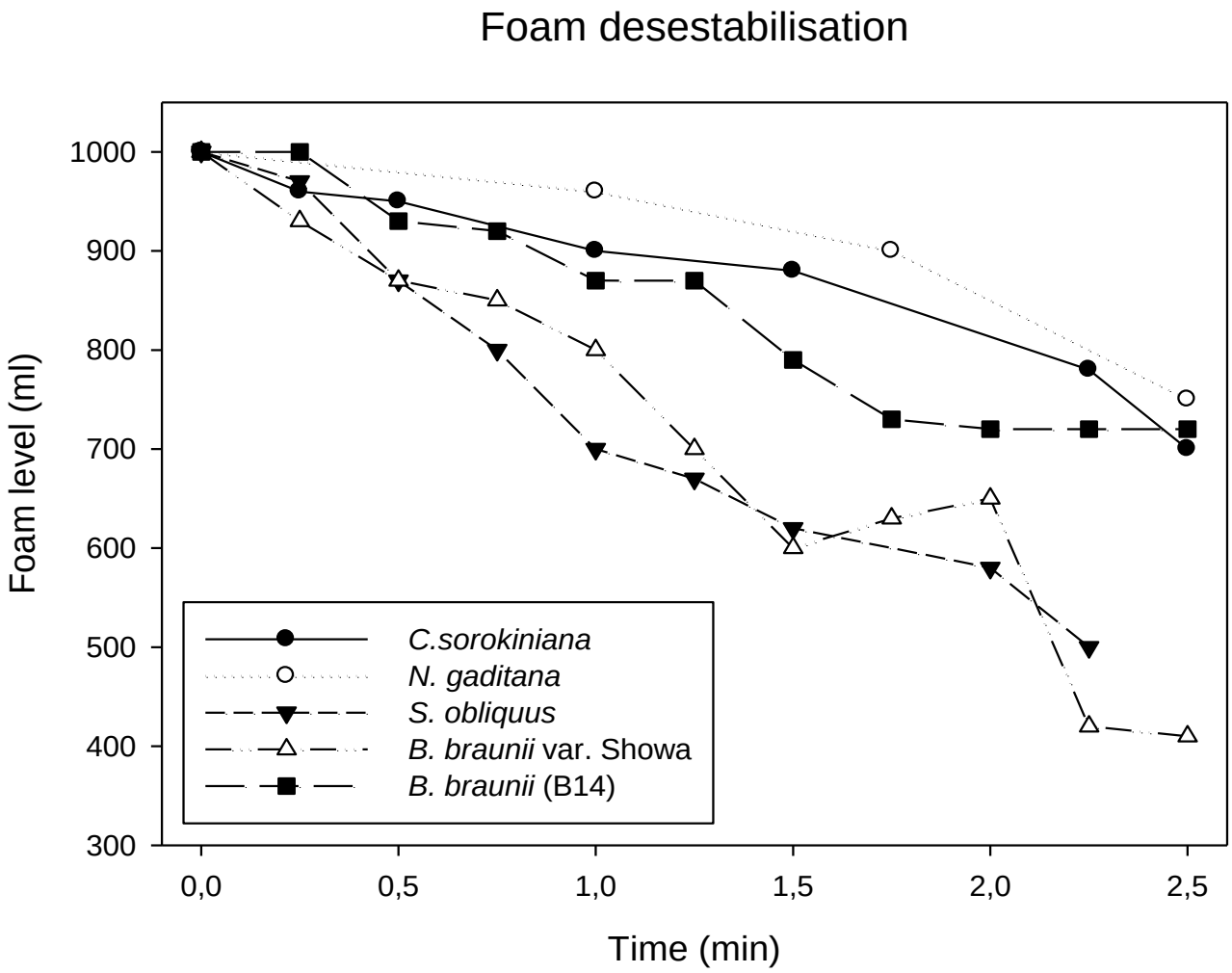
MIRACLES
SPECIALTIES FROM ALGAE

RESULTS

5. Assessment of foam stability



BEST RESULTS:

- ***C. sorokiniana***
- ***N. gaditana***
- ***B. braunii* (B14)**

NOT MEASURABLE:

- ***N. oleoabundans***
- ***I. galbana***

MIRACLES
SPECIALTIES FROM ALGAE

CONCLUSIONS

- For all microalgae tested, the low stability of naturally produced foam prevents its accumulation. Therefore, addition of a surfactant is required to produce microalgae-enriched foam.
- Some microalgae show higher foaming abilities when collected from stationary phase. However, old cultures are not desirable for continuous cultivation.
- Due to low foam stability or low hold-up, *Neochloris oleabundans, Isochrysis galbana and Botryococcus braunii* var. Showa will not be considered for future experiments.
- For exponential microalgal cultures, the most promising results of foam up-flow, microalgae hold-up and foam stability were obtained for *C. sorokiniana, N. gaditana, S. obliquus* and *B. braunii (*B14*).*
- These species will be employed in further studies.

ACKNOWLEDGEMENTS