2023 AOCS Annual Meeting & Expo

Lipid production of microalgae isolated from the Ionian Sea of Greece

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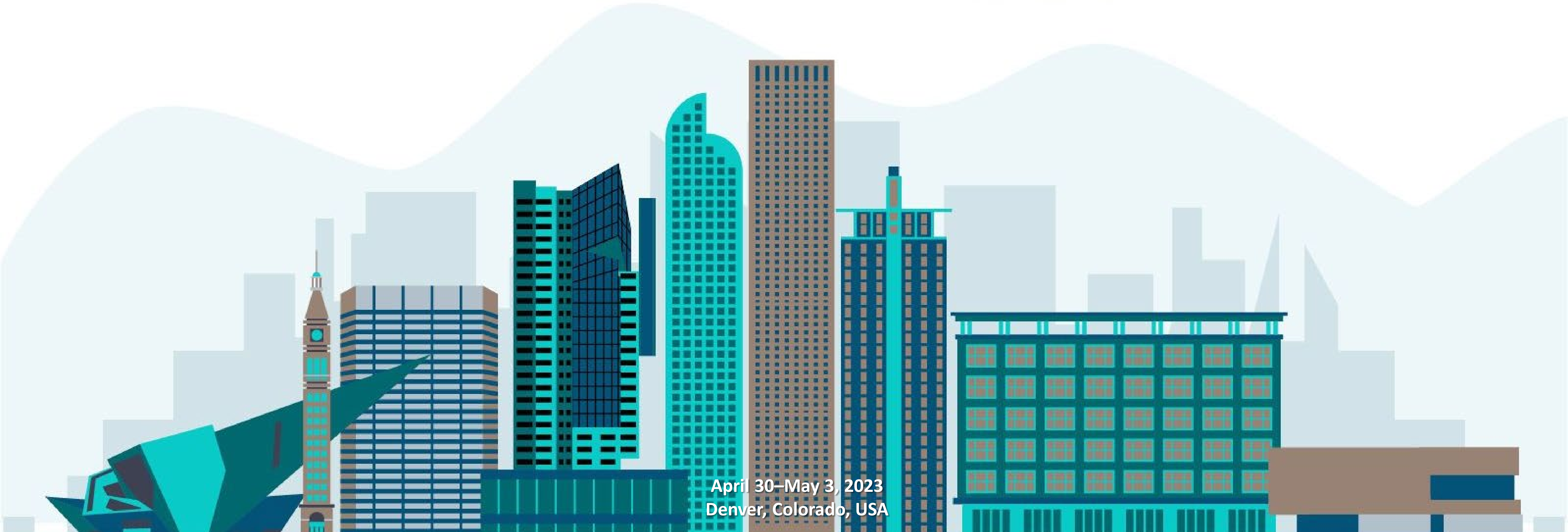
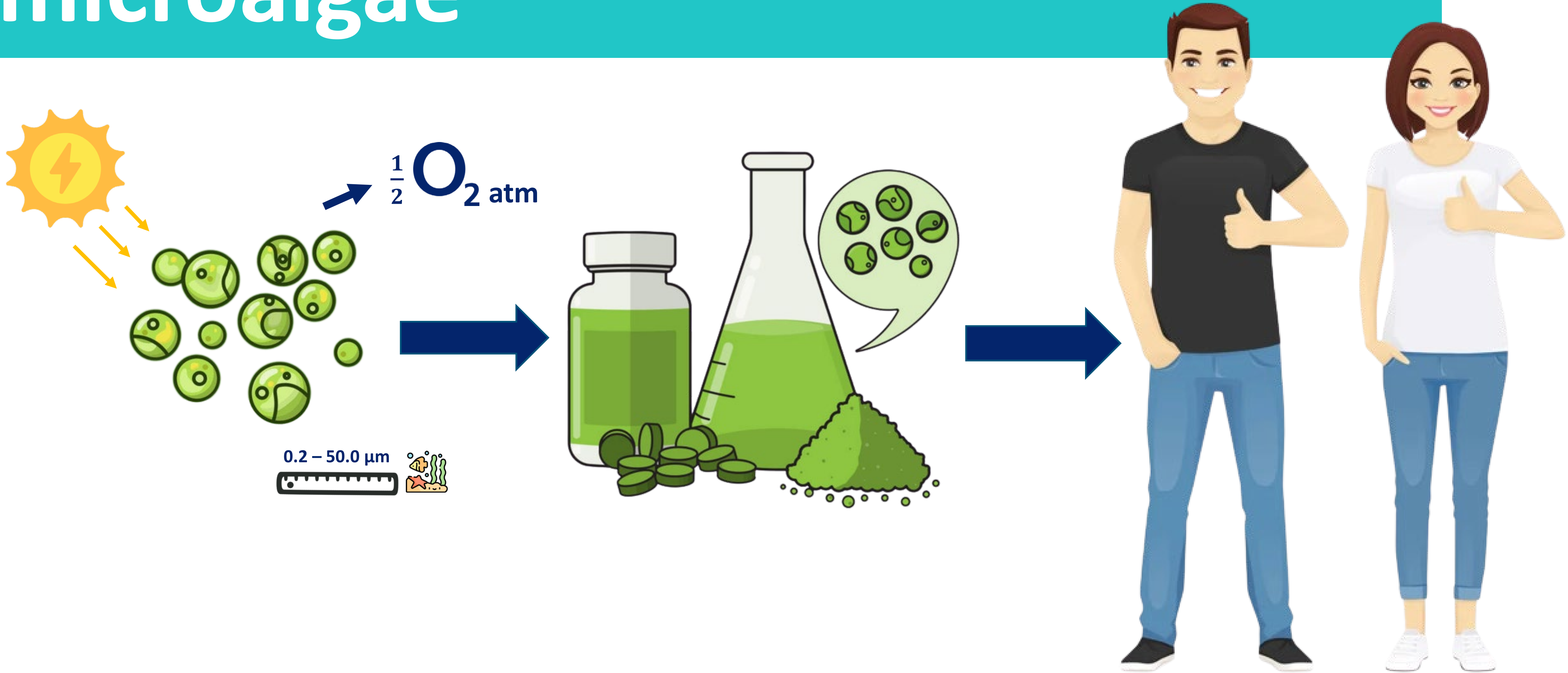
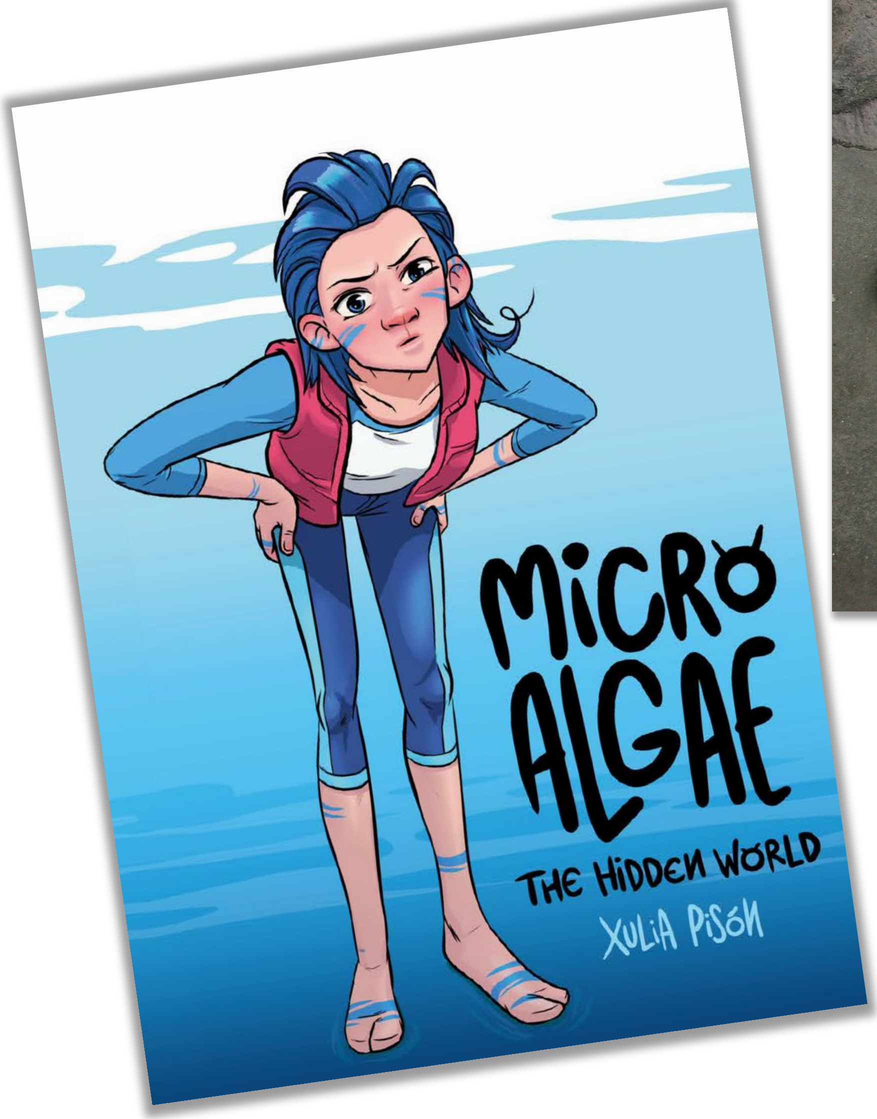
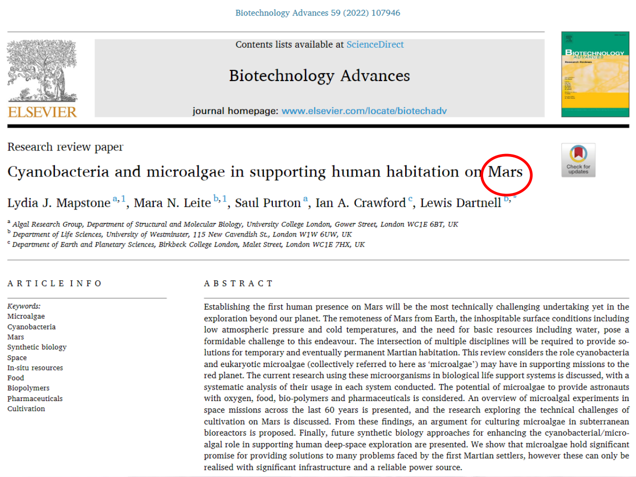


microalgae



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Research review paper

Cyanobacteria and microalgae in supporting human habitation on Mars

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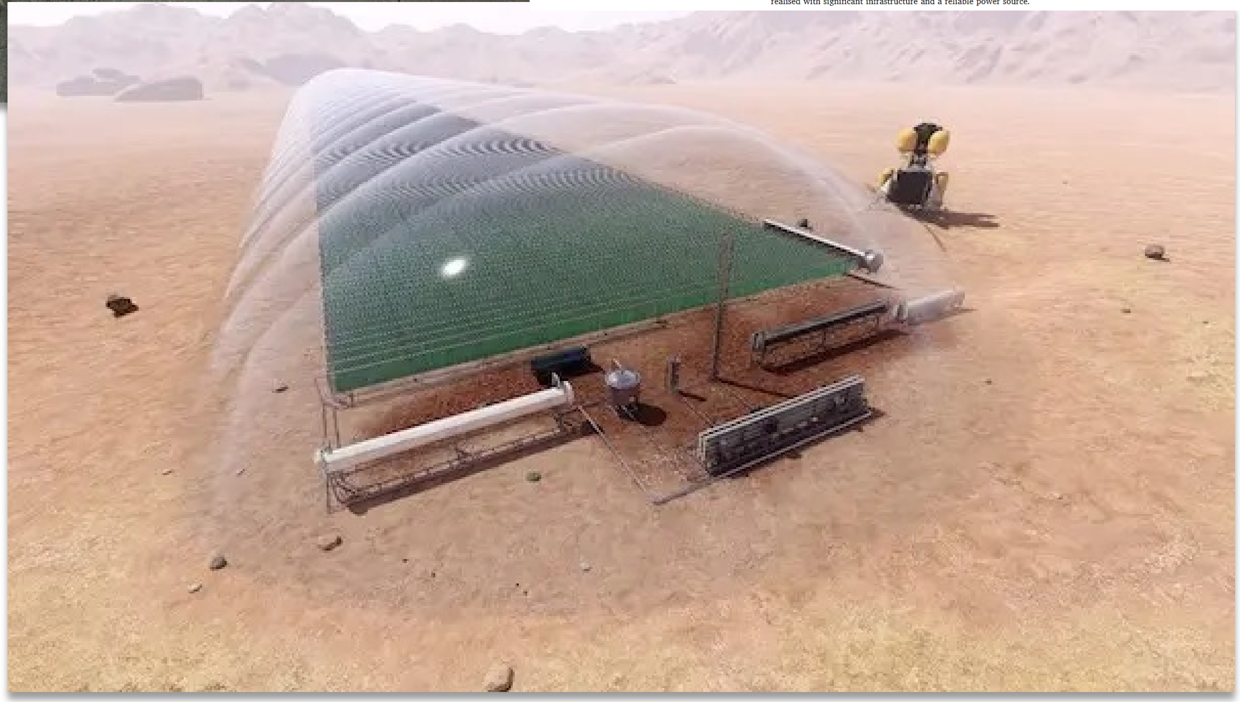
ABSTRACT

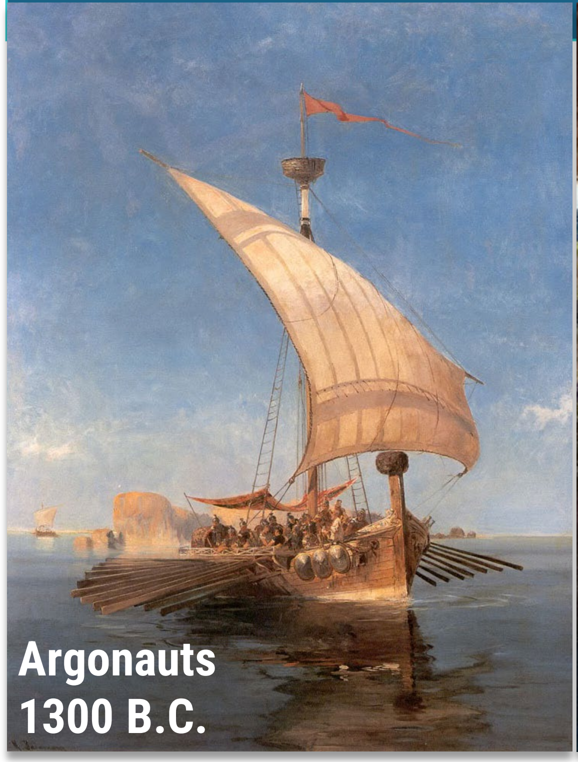
Establishing the first human presence on Mars will be the most technically challenging undertaking yet in the exploration beyond our planet. The remoteness of Mars from Earth, the inhospitable surface conditions including low atmospheric pressure and cold temperatures, and the need for basic resources including water, pose a formidable challenge to this endeavour. The intersection of multiple disciplines will be required to provide solutions for temporary and eventually permanent Martian habitation. This review considers the role cyanobacteria and eukaryotic microalgae (collectively referred to here as 'microalgae') may have in supporting missions to the red planet. The current research using these microorganisms in biological life support systems is discussed, with a systematic analysis of their usage in each system conducted. The potential of microalgae to provide astronauts with oxygen, food, bio-polymers and pharmaceuticals is considered. An overview of microalgal experiments in space missions across the last 60 years is presented, and the research exploring the technical challenges of cultivation on Mars is discussed. From these findings, an argument for culturing microalgae in subterranean bioreactors is proposed. Finally, future synthetic biology approaches for enhancing the cyanobacterial/microalgal role in supporting human deep-space exploration are presented. We show that microalgae hold significant promise for providing solutions to many problems faced by the first Martian settlers, however these can only be realised with significant infrastructure and a reliable power source.

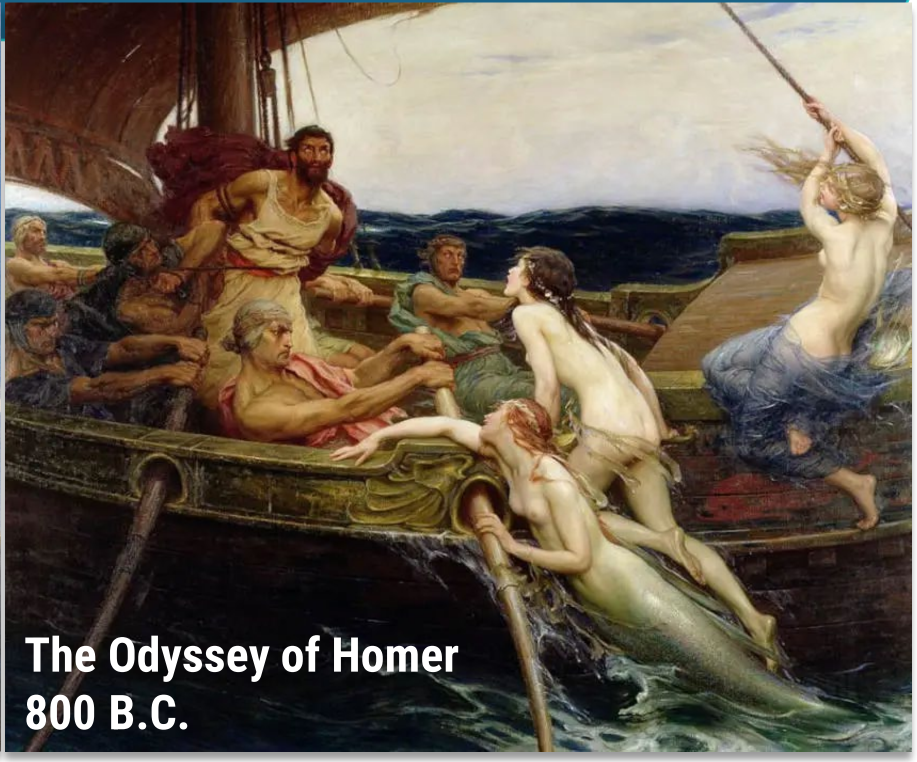


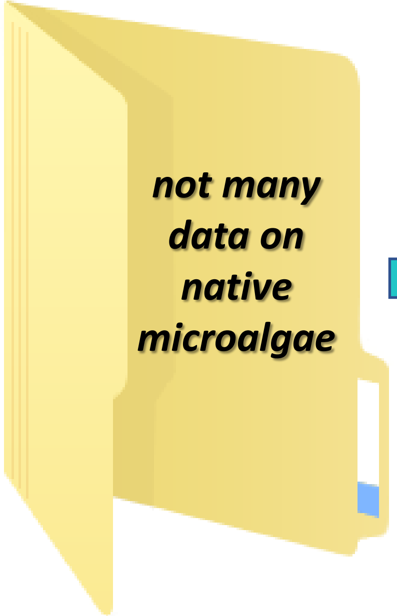
but...

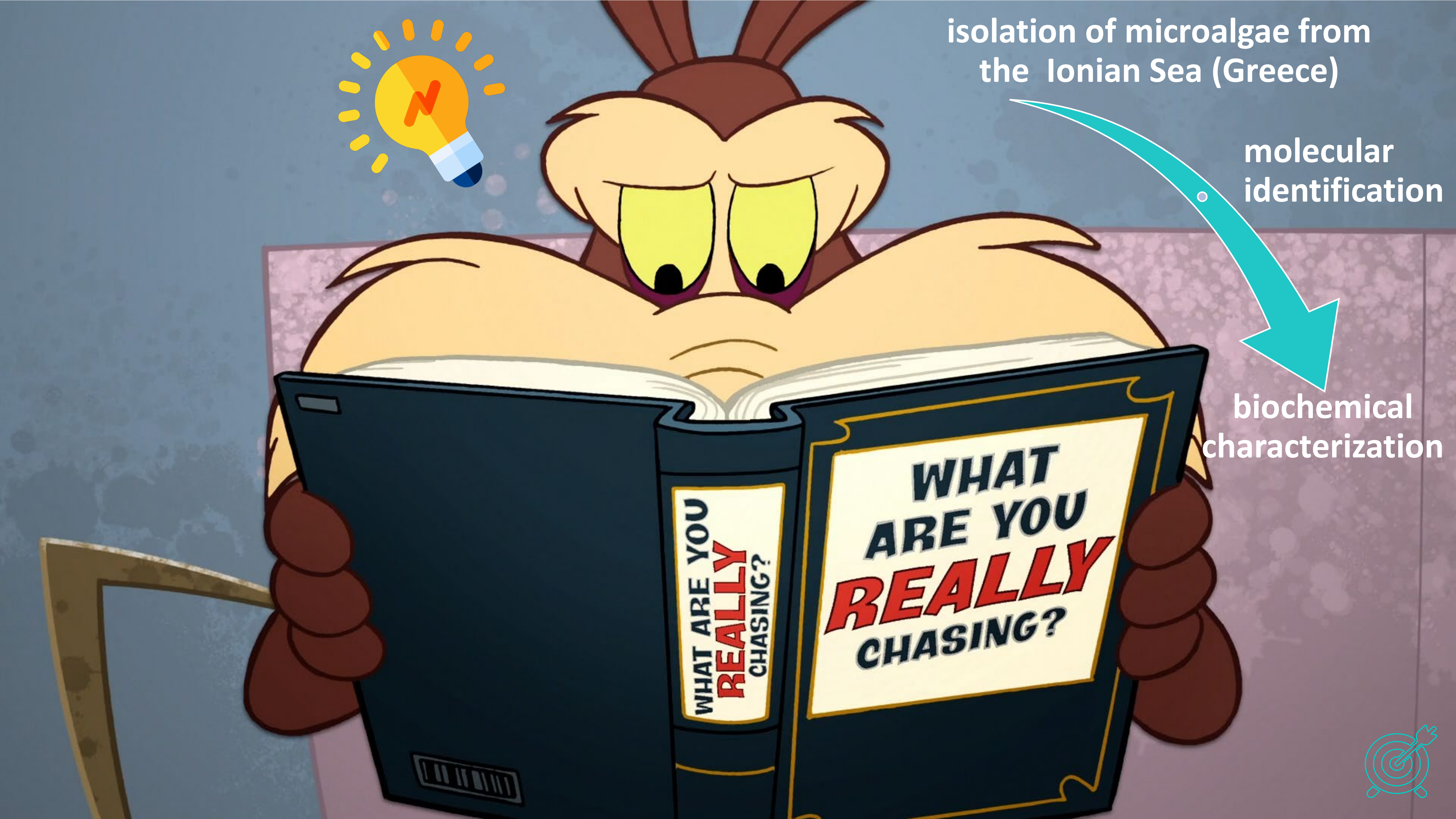
aquaculture in Greece

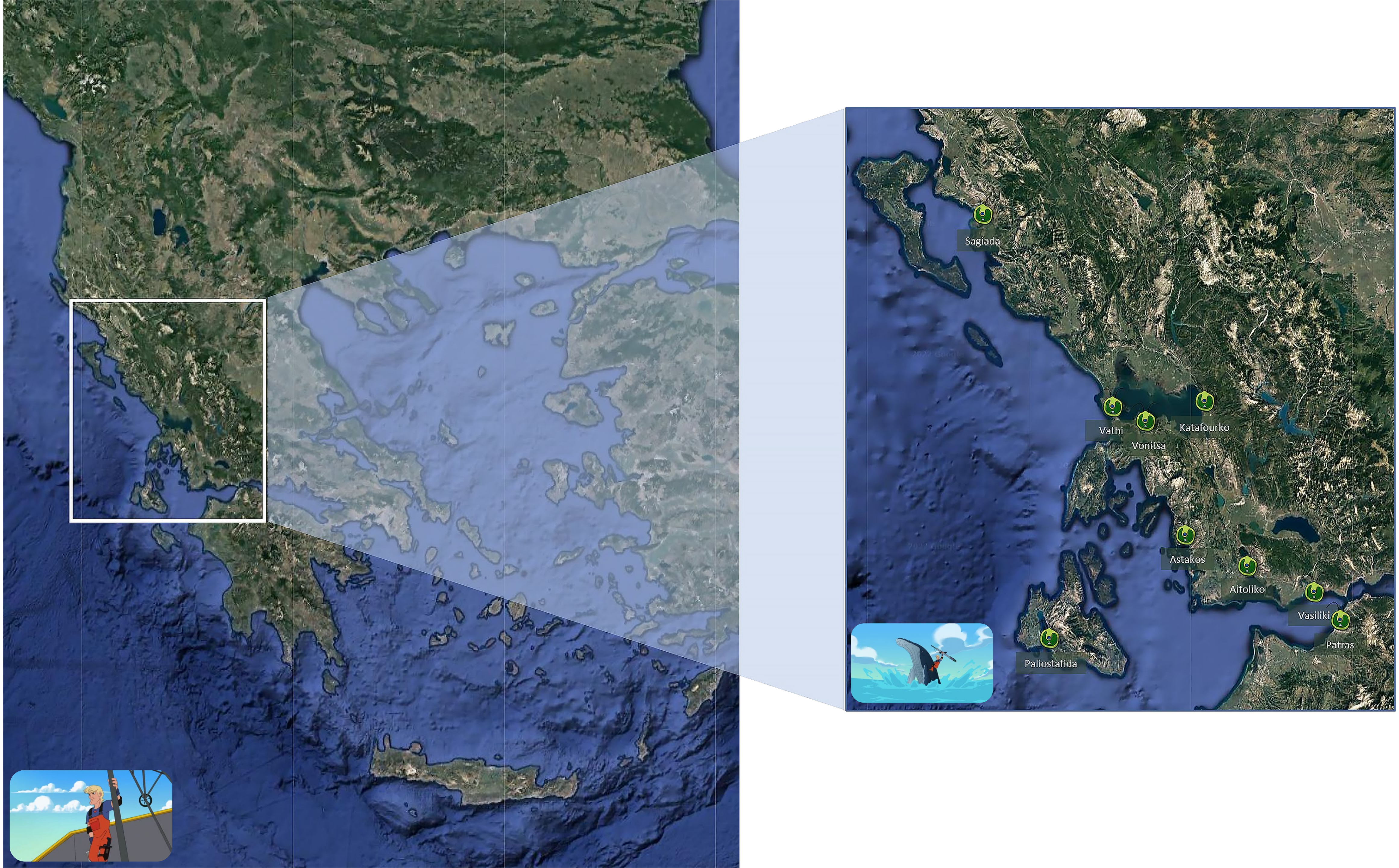
- 61% of fisheries production (FAO, 2019)
- 151,372 tons, worth €589.05 millions → >$600.00 millions
- direct & indirect jobs
- first export sector of livestock production





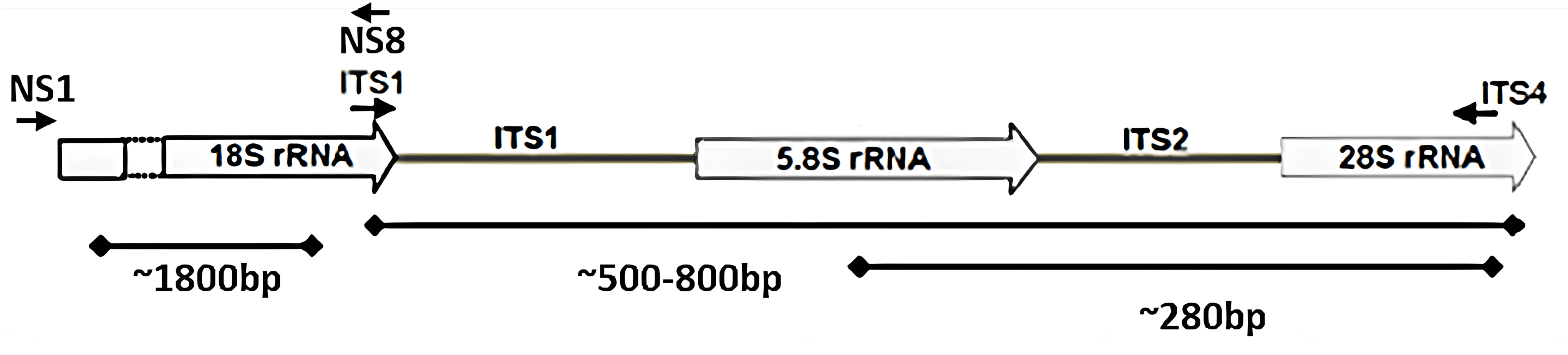



isolation of microalgae from the Ionian Sea (Greece)
molecular identification
biochemical characterization
WHAT ARE YOU REALLY CHASING?
WHAT ARE YOU REALLY CHASING?


Sagiada
Vathi
Vonitsa
Katafourko
Astakos
Aitoliko
Vasiliki
Patras
Paliostafida


ROZAKIS
ΙΑΚΩΒΟΣ
Λ.Γ 1832

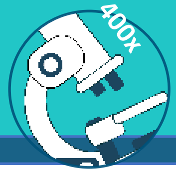
molecular identification

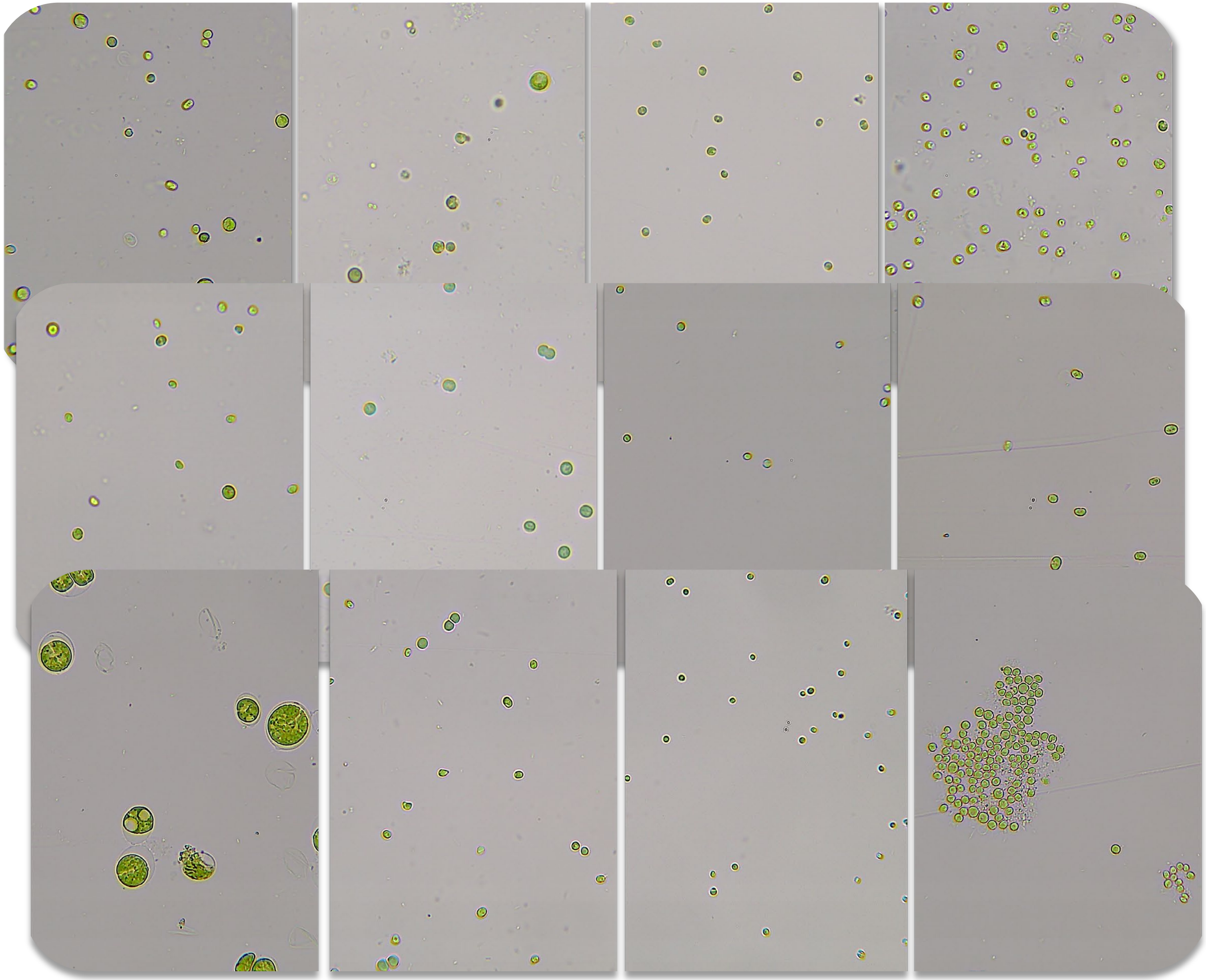


Schematic illustration of the arrangement of the sequences used to identify the microalgae in their genome.

- *Picochlorum*
- *Nannochloropsis*
- *Tetraselmis*
- *Chlorella*
- *Nephroselmis*

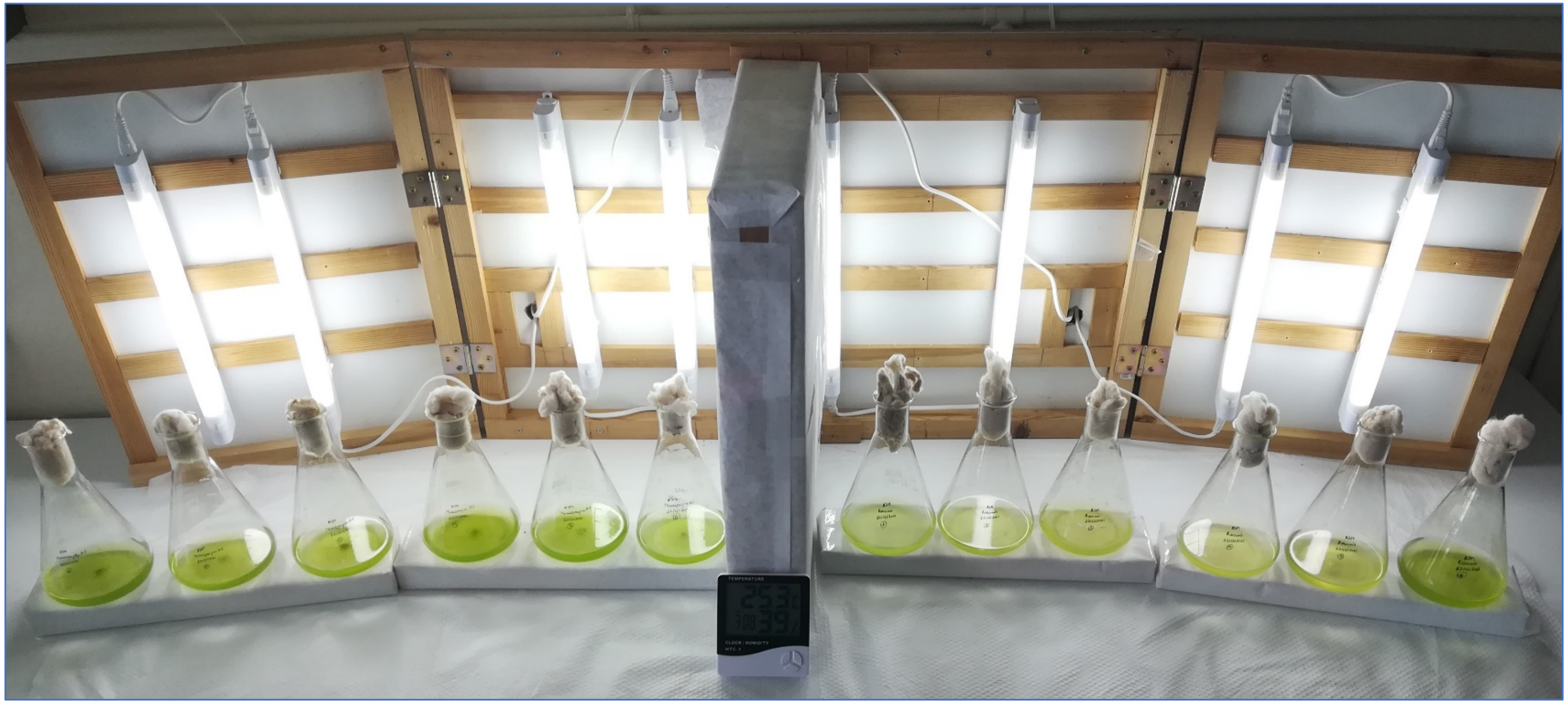
12 new microalgal strains, belonging to 5 different genera





Strain name	Origin	Closest non type species	Non type Accession	Similarity (%)
VAS2.5	Vasiliki. Aetolia-Acarnania	*Picochlorum costavermella*	MT489383.1	96.11%
KAT3.M2	Katafourko. Aetolia-Acarnania	*Tetraselmis* sp.	KC841952.1	99.68%
AIT5.1	Aitoliko. Aetolia-Acarnania	*Nannochloropsis gaditana*	KF040086.1	99.72%
VON5.3	Vonitsa. Aetolia-Acarnania	*Nannochloropsis gaditana*	OM837346.1	98.03%
AST5.2	Astakos. Aetolia-Acarnania	*Chlorella* sp.	X73992.1	98.95%
SAG4.4	Sagiada. Thesprotia	*Picochlorum oklahomense*	AY422073.1	99.94%
VAT4.3	Vathi. Preveza	*Nannochloropsis gaditana*	MN011927.1	99.88%
PAL4.2	Paliostafida. Cephalonia	*Nannochloropsis gaditana*	MN011927.1	99.88%
PATLG-N1	Patras. Achaia (Lighthouse of Patras)	*Nannochloropsis* sp.	MK971790.1; MN011927.1	99.42%
PAT3.2B	Patras. Achaia	*Picochlorum oklahomense*	AY422073.1	99.94%
PAT2.7	Patras. Achaia	*Nephroselmis pyriformis*	EU334587.1	94.79%
PATN2	Patras. Achaia (Port’s Northern Entrance)	*Nannochloropsis* sp.	MK971790.1; MN011927.1	99.82%

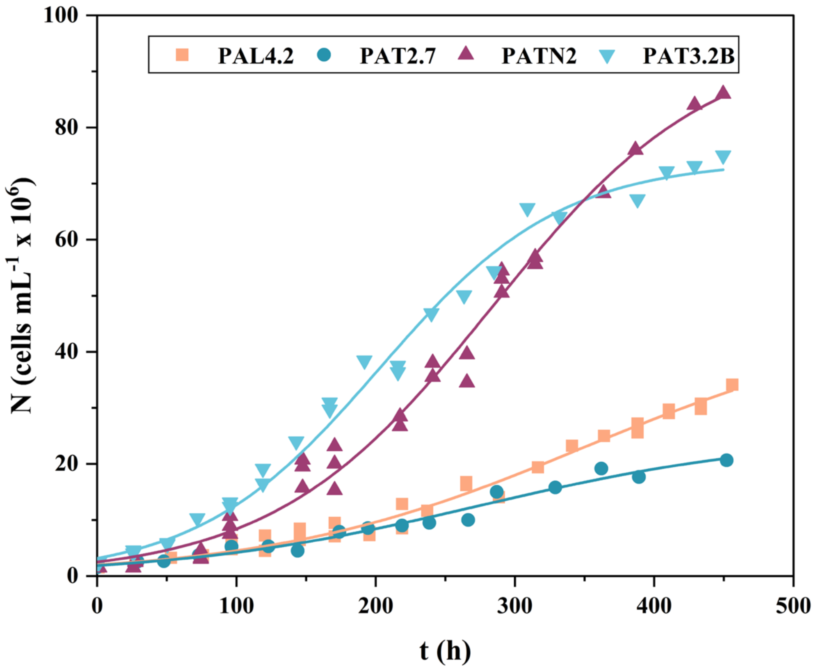
Growth medium: Artificial Sea Water

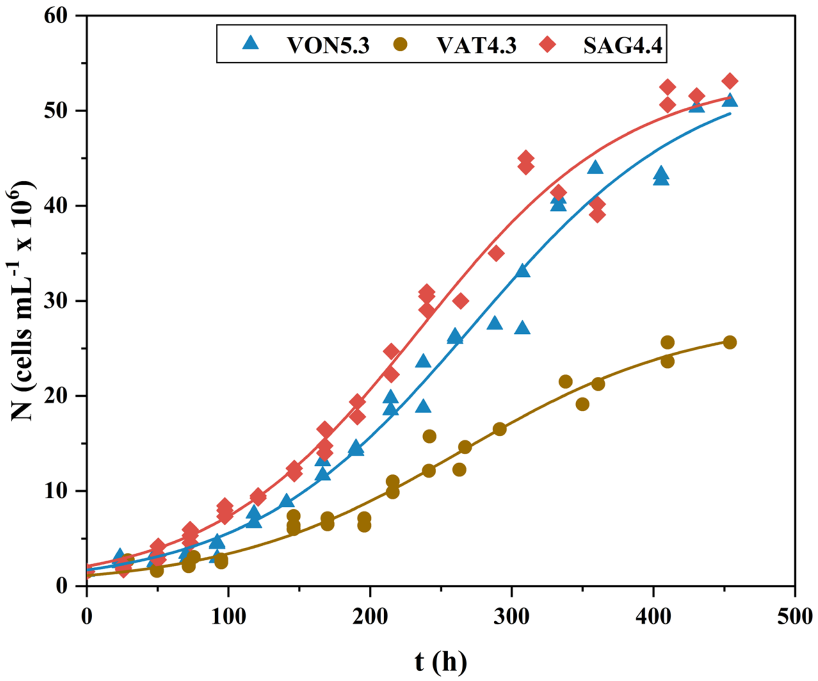


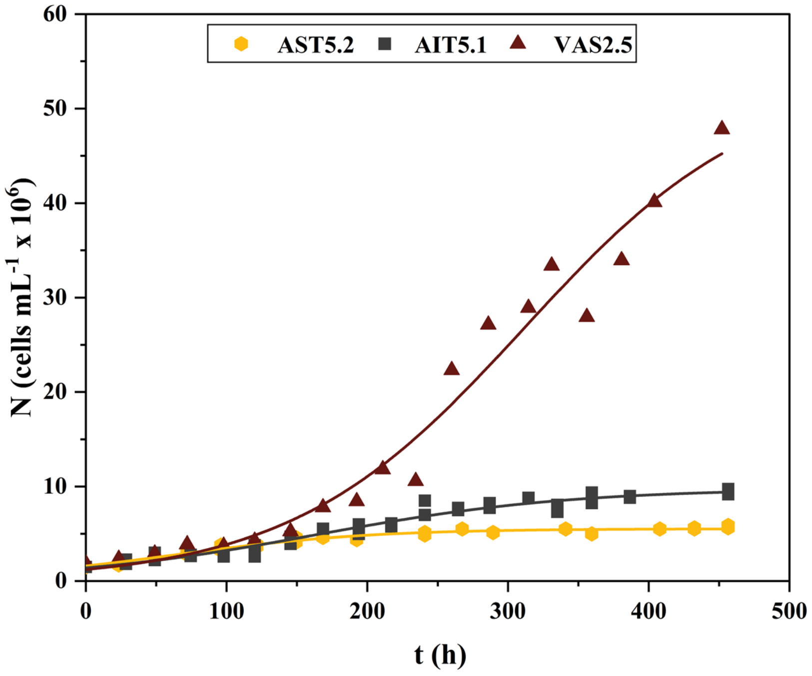
Culture conditions

- **working volume (V_w):** 100 mL
- **inoculum:** 1,5 x 10^6 cells/mL
- **photoperiod:** 24:0
- **illumination:** 387 μmol m^{-2} s^{-1}
- **aeration:** periodical agitation
- **temperature:** 26.0 °C ± 0.5 °C
- **pH:** 8.5 ± 0.5

growth kinetics







adequate growth

all strains, especially those belong to the genera *Picochlorum* and most of *Nannochloropsis*

the highest growth levels

the highest values of μ_{max} were recorded for *Picochlorum* strains (e.g., μ_{max} = 0.37 ± 0.02 d^{-1} achieved by *P. oklahomense* PAT3.2B)

the lowest growth level

Chlorella sp. AST5.2 reaching 6 x 10^6 cells/mL

**Tetraselmis* sp. KAT3.M2 and *Nannochloropsis* sp. PATLG-N1 were excluded due to their tendency to form cell aggregates.

Biomass production, storage materials accumulation and growth parameters from all the cultures that have been carried out.

Strain name	x (mg L^{-1})	L/x (%)	S/x (%)	P/x (%)	N_f (cells mL^{-1} x 10^6)	N_{max} (cells mL^{-1} x 10^6)	μ_{max} (d^{-1})	R^2
P. costavermella VAS2.5	392.1 ± 2.3	**19.1 ± 0.1**	5.0 ± 0.2	**33.0 ± 5.7**	47.8	53.5 ± 6.7	0.29 ± 0.04	0.96
Tetraselmis sp. KAT3.M2	**1558.7 ± 75.2**	**12.8 ± 2.0**	**31.5 ± 0.1**	**57.6 ± 0.7**	ND	ND	ND	ND
N. gaditana AIT5.1	204.6 ± 36.3	**12.8 ± 4.4**	**13.4 ± 1.1**	19.2 ± 2.7	9.8	9.8 ± 0.4	0.26 ± 0.02	0.96
N. gaditana VON5.3	**473.1 ± 30.1**	**9.3 ± 3.0**	7.4 ± 0.1	26.7 ± 0.9	50.9	54.7 ± 2.3	0.30 ± 0.02	0.98
Chlorella sp. AST5.2	188.3 ± 9.9	3.4 ± 0.6	**13.7 ± 1.2**	29.3 ± 1.4	5.6	5.5 ± 0.1	0.34 ± 0.03	0.96
P. oklahomense SAG4.4	285.8 ± 6.2	8.6 ± 1.1	7.7 ± 1.7	26.1 ± 4.4	53.1	53.9 ± 1.7	0.33 ± 0.02	0.98
N. gaditana VAT4.3	**513.7 ± 39.3**	**10.4 ± 0.6**	7.4 ± 0.8	16.5 ± 1.9	25.6	28.4 ± 1.5	0.29 ± 0.02	0.98
N. gaditana PAL4.2	**636.9 ± 95.3**	7.2 ± 1.2	6.1 ± 0.8	**31.1 ± 3.1**	34.1	46.0 ± 4.2	0.21 ± 0.01	0.99
Nannochloropsis sp. PATLG-N1	**639.4 ± 22.8**	**11.1 ± 1.2**	10.4 ± 0.0	21.9 ± 0.7	ND	ND	ND	ND
P. oklahomense PAT3.2B	320.3 ± 55.1	**10.3 ± 2.2**	7.2 ± 2.1	**43.4 ± 1.4**	75.0	74.1 ± 1.4	0.37 ± 0.02	0.99
N. pyriformis PAT2.7	**628.5 ± 56.2**	4.2 ± 1.2	**13.4 ± 1.6**	**46.8 ± 2.4**	20.6	25.1 ± 2.8	0.22 ± 0.03	0.97
Nannochloropsis sp. PATN2	**959.5 ± 11.2**	**11.1 ± 0.0**	9.1 ± 0.1	22.5 ± 5.8	86.0	95.8 ± 3.3	0.31 ± 0.01	0.99

Fatty acid composition of total lipid of the isolated microalgae strains at the end of their culture in 0.5-L Erlenmeyer flasks (V_w = 0.1 L). Experiments were performed in duplicate.

Strain name	Fatty acid composition (w/w%) of total lipid												
	C14:0	$^{\Delta 9}$C14:1	C16:0	$^{\Delta 9}$C16:1	C17:0	C18:0	$^{\Delta 9}$C18:1	$^{\Delta 9,12}$C18:2	$^{\Delta 9,12,15}$C18:3	$^{\Delta 6,9,12,15}$C18:4	$^{\Delta 13}$C20:1	$^{\Delta 5,8,11,14,17}$C20:5	*Others
P. costavermella VAS2.5	8.9	6.2	28.5	38.9	0.8	1.0	10.1	2.3	0.2	**2.3**	ND	ND	0.9
Tetraselmis sp. KAT3.M2	1.6	2.1	21.9	10.1	1.4	6.2	27.4	5.4	**11.9**	**4.1**	2.7	2.8	2.4
N. gaditana AIT5.1	1.1	5.4	18.7	10.3	ND	1.7	15.1	15.0	**15.4**	ND	< 0.1	5.1	12.2
N. gaditana VON5.3	8.5	3.4	19.8	30.6	ND	1.6	6.6	0.7	0.2	0.2	0.6	**24.3**	3.5
Chlorella sp. AST5.2	0.4	1.8	15.2	2.9	12.8	1.9	16.9	13.7	**29.8**	ND	ND	ND	4.5
P. oklahomense SAG4.4	1.2	5.1	15.9	7.3	5.6	1.9	27.0	10.2	**19.6**	< 0.1	ND	ND	6.3
N. gaditana VAT4.3	4.7	1.9	27.7	28.6	0.3	1.1	8.5	2.7	ND	ND	4.3	**17.3**	3.1
N. gaditana PAL4.2	7.3	3.2	20.2	27.8	0.6	0.7	7.4	3.2	ND	ND	2.5	**23.4**	3.7
Nannochloropsis sp. PATLG-N1	2.9	1.4	26.1	40.2	ND	0.3	10.5	1.4	ND	ND	3.5	10.7	3.0
P. oklahomense PAT3.2B	0.9	5.6	13.1	5.1	4.9	1.4	22.1	16.4	**18.2**	**2.8**	ND	< 0.1	9.6
N. pyriformis PAT2.7	28.8	4.8	9.3	38.8	0.4	4.5	7.0	0.7	ND	ND	ND	ND	5.8
Nannochloropsis sp. PATN2	3.6	1.1	26.4	33.2	ND	2.0	13.7	3.0	0.1	ND	3.0	11.8	2.1

Abbreviations: ND, not detected
***Others:** C10:0, C12:0, C16:2, $^{\Delta 6,9,12}$C18:3

Conclusions

Picochlorum, *Tetraselmis*, *Nannochloropsis*
biomass & high amounts of proteins & lipids rich in PUFAs

suitable for **aquaculture or** various applications in the food, feed and pharmaceutical **industries**

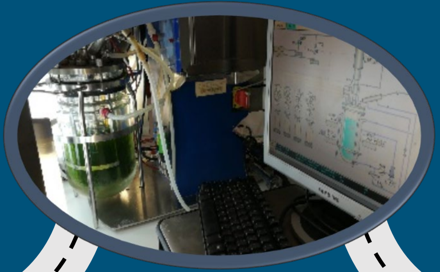
Tetraselmis sp., *N. pyriformis* & *Chlorella* sp.
polysaccharides → bioethanol production

higher levels synthesis of these metabolites **compared to commercial strains**

Coming next...

cultivation of selected microalgae in V_w = 2000 mL

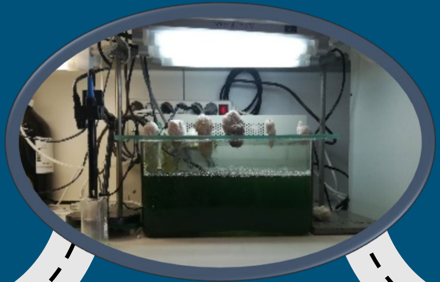
1



effect of temperature

cultivation of selected microalgae in V_w = 5000 mL

2



effect of nitrogen, phosphorus, CO_2, pH, photoperiod

mixotrophic cultivation

3

Acknowledgements

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our team


Panagiotis Dritsas


Elias Asimakis


George Tsiamis


George Aggelis


PRESENTATION IS FINISHED
THANK YOU
FOR YOUR ATTENTION!!!