Netzwerk Algen 2018
Kultivierungsbedingungen und Kontamination
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Cultivation of Microalgae

Photobioreactors and Contamination

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Outline

- ALGATECH Centre - Microalgae Biotechnology in Třeboň
- Cultivation systems
- Contamination - Anti-contamination strategies

PROJECTS

- EU H2020 SABANA project (2017-2020)
- Interreg project ALGENETICS

About ALGATECH Centre – since 2011

>100 employees

Microalgal Biotechnology since 1960s

Laboratories:

Photosynthesis / Cell Cycles / Photosynthetic Bacteria

Algal Biotechnology

Topics in Microalgae Research

Physiology / **Photosynthesis** / Molecular Biology / Biophysics / Biochemistry / Cell Biology / **Biotechnology**

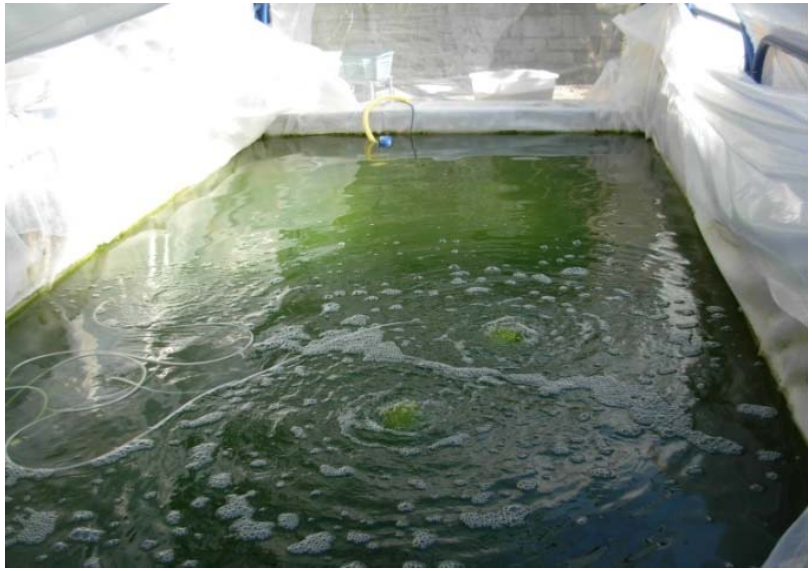
Cultivation Systems

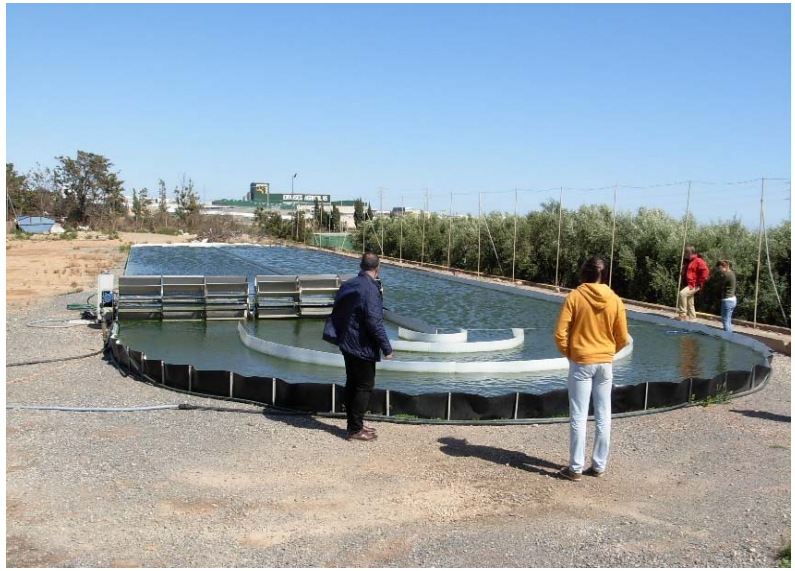
TWO TYPES

Open systems
Ponds, raceways
cascades

Closed or semi-closed
systems = PBRs

Open Production Systems for Microalgae Cultivation





Outdoor PBRs for Microalgae Cultivation

Greenhouse PBRs or Microalgae Cultivation

Maintenance, cleaning, contamination

OPEN PONDS RACEWAYS CASCADES	CLOSED OR SEMI-CLOSED PBRs
o Cheap construction and maintenance	o Higher costs for construction and maintenance
o Cleaning easy	o Cleaning !!!
o Low control of culture conditions	o Good control of culture conditions
o Evaporation – cooling !!!	o Cooling !!!
o Contamination !!!	o Low Contamination

Strain selection

OPEN PONDS RACEWAYS CASCADES	CLOSED OR SEMI-CLOSED SYSTEMS=PBRs
o **Species specific** o Fast-growing, robust strains – *Chlorella, Scedesmus*, etc. (*Chlorophyta*) o Specific conditions - *Spirulina* (bicarbonate , high pH), *Dunaliella* (high salinity), marine *Nannochloropsis*	o **Spectrum of species** o Production strains – *Haematococcus, Chlorella mixotrophic* o *Strains for high-requirements* o *Aquaculture strains – Isochrysis, Tetraselmis, Phaeodactylum, Skeletoma* o Cultivation of GMO

Contamination / Infection in Microalgae Cultures

- **Chemical** – heavy metals, PCB, PAH – clean water, chemicals
- **Unwanted microalgal strains** (mostly fast growing *Chlorophyta – Chlorella, Scenedesmus, Selenastrum*) – cleaning inoculation from monocultures
- **Fungi** (*Chytridium, Aphelidium*)
- **Bacteria** – non-smooth surfaces harbour biofilms, organic matter residues (steam treatment, mechanical cleaning)
- "**Friendly" bacteria** (*Micrococcus, Bacillus*) – not abundant in inorganic media/phototrophic cultures – growth in stressed cultures (can be used for feed and food $<10^5$ CFU/mL
- **Pathogenic bacteria** – *Escherichia (coliforms), Salmonella, Staphylococcus, Clostridium*
- **Predators** - freshwater phytoplankton *Vorticela* (Protozoa), golden algae *Poterioochromonas* (Synurophyceae)
- **Toxic cyanobacteria** - *Microcystis, Anabaena*
- **Zooplankton -** rotifers, etc. – spread by birds
- **Viruses, phages**

Anti-contamination Strategies

- **Case specific -** when infected: harvesting and cleaning/sanitation, → new inoculation from monoculture
- **Favourable growth conditions** – contamination less probable in well-growing 'aerobic' cultures – defense mechanisms
- **Regular observation of culture** – microscopic, colour, smell, flocculation – sedimentation, appearance, decrease of photosynthetic activity (DOC), etc.
- **Regular cleaning** (daily TLCs)
- **Disinfection** – hypochlorite (H-O-Cl, production of chlorine), Dosyl (H_3PO_4, $H_2SO_4 + H_2O_2 \rightarrow O_3$)
- **UV light, ozone treatment –** system, medium
- **Specific environmental conditions** – high-salt, high pH
- **High concentration of inoculum** - monocultures
- **Use of fungicides and antibiotics** – problematic

Open Outdoor Systems – Contamination

- ***Vorticella* contamination** *(protozoa, phylum Ciliophora)*
- Appear when very high growt rate (high temperature >38 °C, high irradiance) → change of colour, smell

Action – higher flow rate, decrease of pH < 3

Contamination in PBRs - Synechocystis vs. Poterioochromonas

- Synechocystis predating species of **golden algae** ***Poterioochromonas***
- **Defense strategy** against protozoa and other competing microalgae**:** **high pH >11**

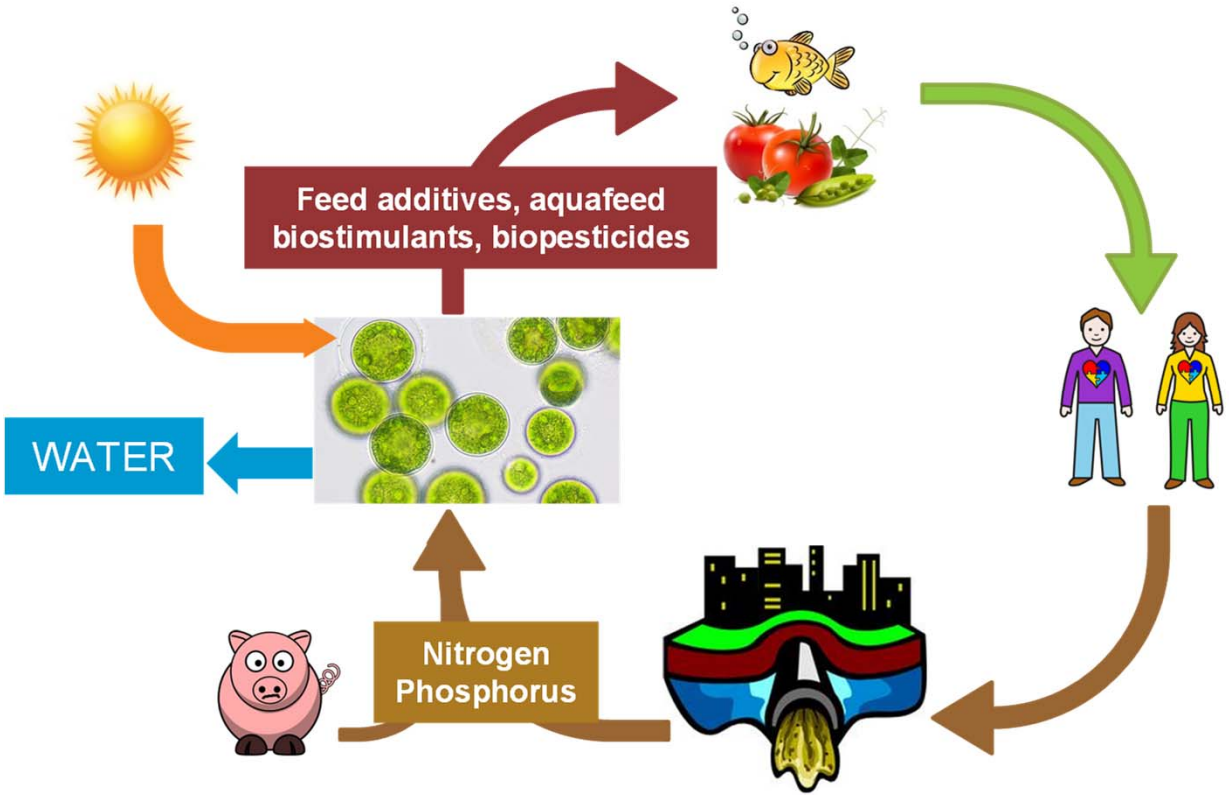
Case Studies – Microalgae in Agriculture

Sustainable Integrated **A**lgae **B**iorefinery for the Production of Bioactive Compounds for **A**griculture a**N**d **A**guaculture

EU H2020 Research and Innovation Programme (2016-2020)

Coordinator: University of Almería, Spain (prof. Gabriel Acién)

Block Diagram of SABANA Project



Task of ALGATECH team

- Design and construction of cultivation units (Algatech, University of Almeria)
- Characterisation of selected microalgae strains
- Development of monitoring techniques for large scale system

ALGENETICS

Joint Czech - Austrian Centre for Algal Biotechnology

INTERREG – Cross-border collaborative program Austria-Czech Republic (Upper Austria and South Bohemia)

- FH OÖ Forschungs & Entwicklungs GmbH in Wels
- Centre Algatech, Institute of Microbiology in Třeboň

Strategic Partners:

- FH OÖ Studienbetriebs GmbH in Wels
- University of South Bohemia in České Budějovice

ALGENETICS

- Construction, selection and characterisation of genetically modified cyanobacteria (over)producing glycogen and starch
- Optimising cultivation regimes of mutants from laboratory to pilot scale cultivation

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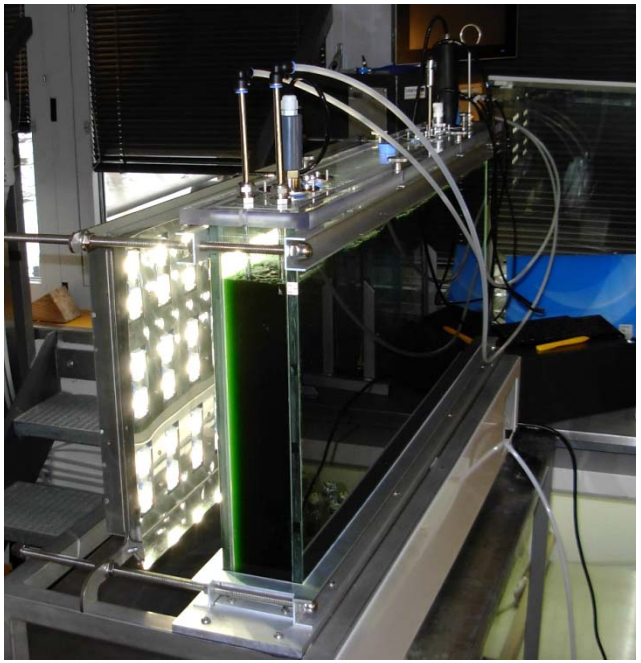
- Closed systems – photobioreactors → work with GMO

Laboratory cultivation of *Synechocystis* PCC6803

10-L PBR



25-L fully controlled
Flat Panel PBR

100-L PBR
with internal illumination

Potential production of Starch by Cyanobacteria

Supplement Production of EtOH from wheat Starch

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Thank you for attention

Questions,
remarks?

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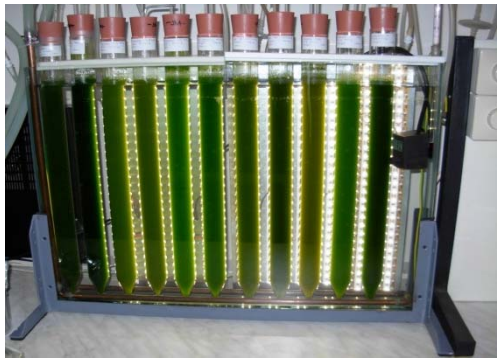
From Collection to Small Scale Culture

1st – collection of strains on agar



2nd step – 400 mL