Centre ALGATECH
Institute of Microbiology
Třeboň, Czech Republic

Microalgal biotechnology in the Centre Algatech in Trebon. Overview of mass cultivation and downstreaming

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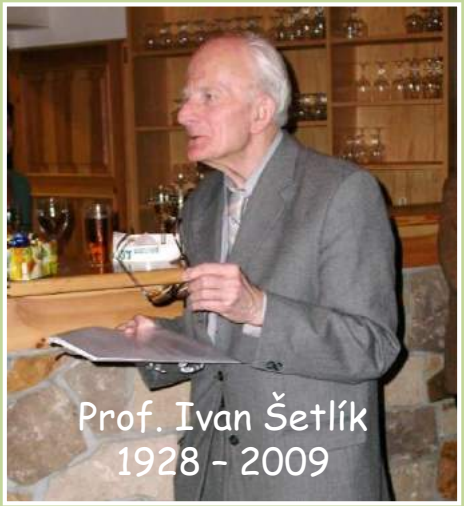
Past and present: a timeline Microalgal Biotechnology in Czechoslovakia (Czech Republic)

Large scale cultivation of microalgae Institute of Microbiology Třeboň, Czechoslovakia


Prof. Ivan Šetlík
1928 – 2009

1940
1950
1960
1970
1980
1990
2000
2010

Proposal to produce oil from algae (diatoms) (Germany)

Algal culture: from laboratory to pilot plant (Burlew 1953)

Chlorella production (Japan, Taiwan)

Single cell protein from microalgae
Wastewater treatment with algae (USA, Germany, Israel)
Spirulina production (Mexico)

Dunaliella production (Australia, Israel, USA)

Microalgae for liquid fuels (SERI, USA)
Haematococcus production (USA)

CO_2 bioremediation (RITE, Japan)

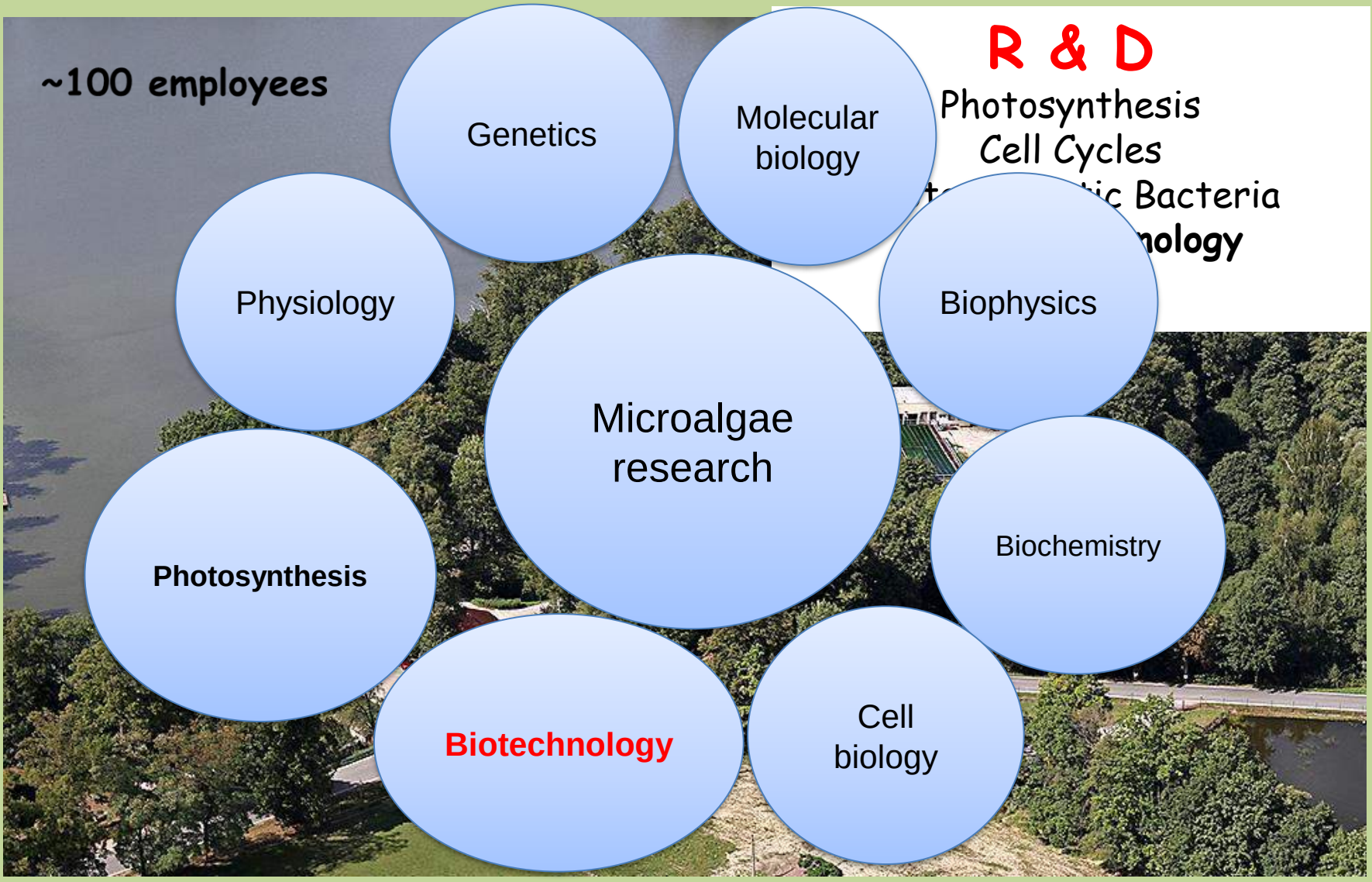
'Rediscovery' of microalgae for biofuels

1960s Inclined platforms with baffles – first large-scale systém in Europe

Unique thin-layer cultivation system – highly productive, high cell density cultures designed in the early 1960s by I.Šetlík & co-workers for growth of microalgae Institute of Microbiology, Academy of Science, Třeboň, 900 m²

Šetlík et al. (1970) Dual purpose open cultivation units for large scale culture of algae in temperate zones. Algological Studies 1: 111-164.

Centre for Algal Biotechnology - Algatech Třeboň, est. 2011



Research topics

Laboratory of Algal Biotechnology

- Screening and selection of microalgae strains
- Design and construction of various cultivation units
- Optimisation of culturing regimes for microalgae
- Heterotrophic cultivation of microalgae
- Production of biomass as food and feed additives, for isolation of valuable compounds, etc.
- Identification and characterisation of bioactive compounds with potential pharmacological use – analytical techniques

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Process

USP

DSP

slant agar

Product

Advantages of heterotrophy

- controlled cultivation conditions
- no weather influence
- little space necessity
- high yields
- simple scale-up

- ability to direct produce at pharma grade
- closed systems for save GMO cultivation

Disadvantages heterotrophy

- limited group of strains
- ability vs. interest
- easy contamination with other heterotrophs
- few axenic strains in culture collections
- difficult screening
- carbon footprint cultivations

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USP

- lab inoculum preparation
- F150 inoculum cultivation
- F1500 inoculation
- F1500 production batch cultivation

18 L

150 L

1500 L

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DSP

- Biomass harvesting and dilution
- Centrifugation
- Disintegration
- Spray drying

Counter current Chromatography (CCC)

• CCC - liquid/liquid chromatography technique (two immiscible liquids)

• One liquid phase (the stationary phase) is retained in the column by centrifugal force, second (the mobile phase) is pumped through the column.

Benefits of CCC

- ✓ No risk of irreversible adsorption of analytes *(*no solid support).
- ✓ High loading capacity of sample (high throughput).
- ✓ Cost-effective technique (low solvent consumption).
- ✓ Greener technology (non-toxic and biodegradable solvents).
- ✓ Scalable - from laboratory to pilot and industrial size.
- ✓ Automated operation and fully predictable separation.
- ✓ High recovery of injected sample (liquid stationary phase).
- ✓ Multifunctional technique (extraction, fractionation, isolation, purification).

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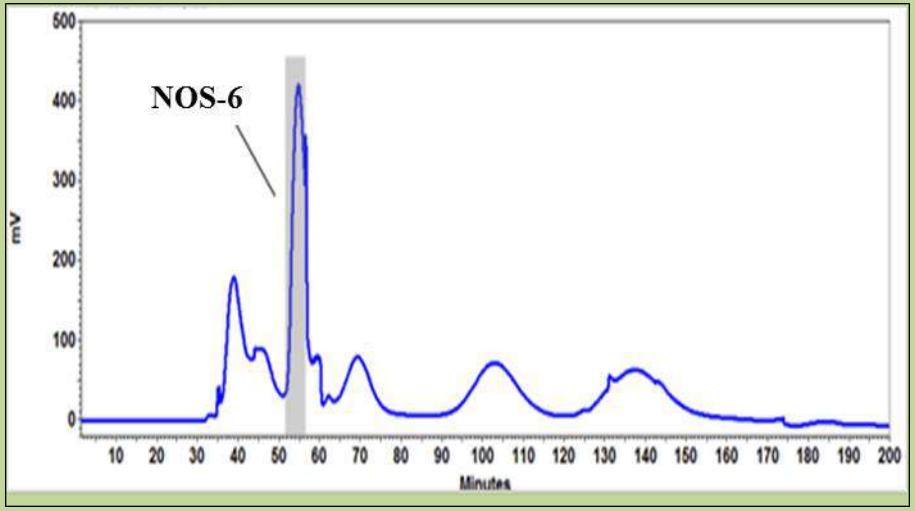
HPCCC separation of cold acetone-treated extract of *Nostoc* sp.20

HPCCC parameters	
Column volume	134 mL (Dynamic Extractions - Spectrum HPCCC)
Solvent System	*n*-hexane–ethyl acetate–MeOH–water (HEMWat, 4:5:4:5, v/v/v/v)
Rotational speed	1000 rpm
Flow rate	1 mL/min
Stationary phase	83%
Run Mode	Reverse, Isocratic
Injection Volume	3 mL
Sample	100 mg acetone-treated crude extract in 3 ml (UP/LP, 1:1)
Detection	280 nm



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Scale-up

Traditional way

Test the viability of the big process in lab scale.

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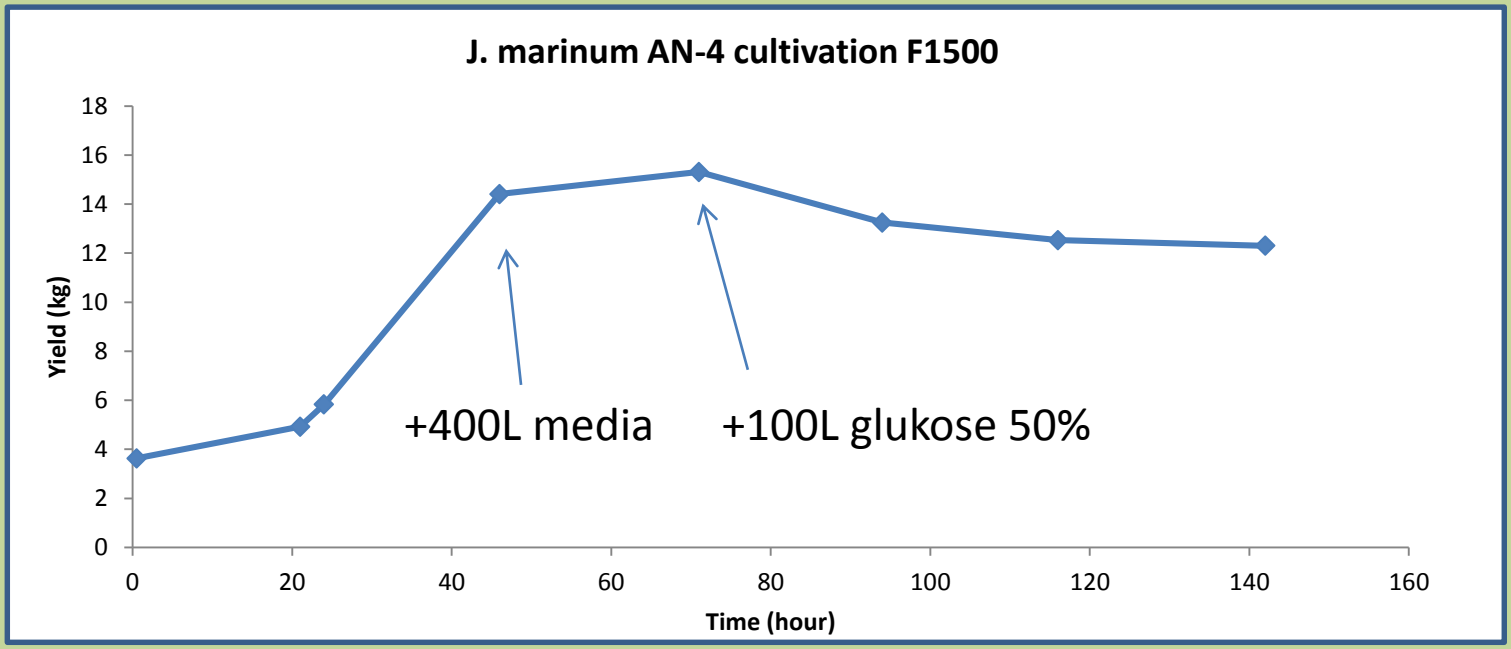
Cultivation *J. marinum*

Inoculation: 16-18L dense culture

Cultivation:

two steps: 1. grow 27 °C

2. lipid production 23-24 °C



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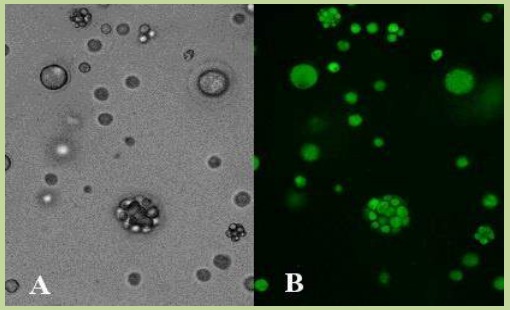
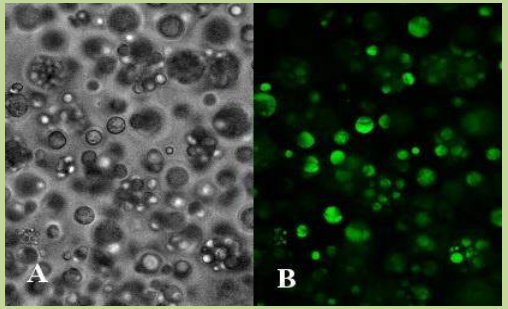
DSP

Lamellar centrifuge EVODOS 10
- fixed rpm 1400
- feed regulation 700L/hour

separation efficiency 51%



End sample F1500



Spray dryed sample F1500

Lipid extract

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Sumary results

Batch	FA (g/kg DW)	DHA mg/L per day
1.	33.7	60
2.	108.8	205
3.	173.64	260

Vessel F1500 before harvesting

Day	FA (g/kg)	DHA (mg/kg DW)
4	87.21	53.07
5	104.07	59.07
6	173.64	92.02

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J.marinum AN4 spray dried

Chlorella vulgaris BEIJ/H14

Thank you for attention