

# Cianobatteri e fissazione biologica dell'azoto per un'agricoltura sostenibile: le prospettive per biofertilizzanti e biostimolanti

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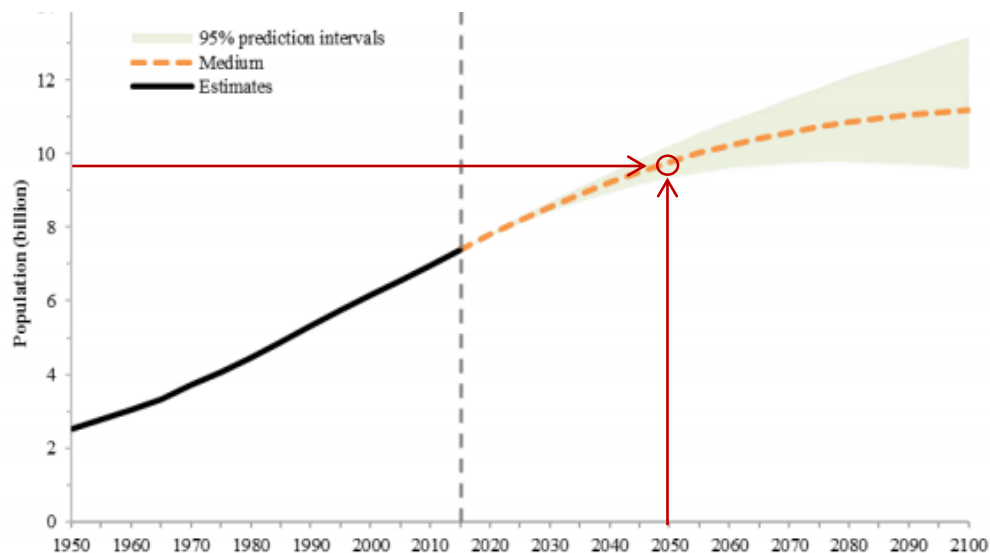
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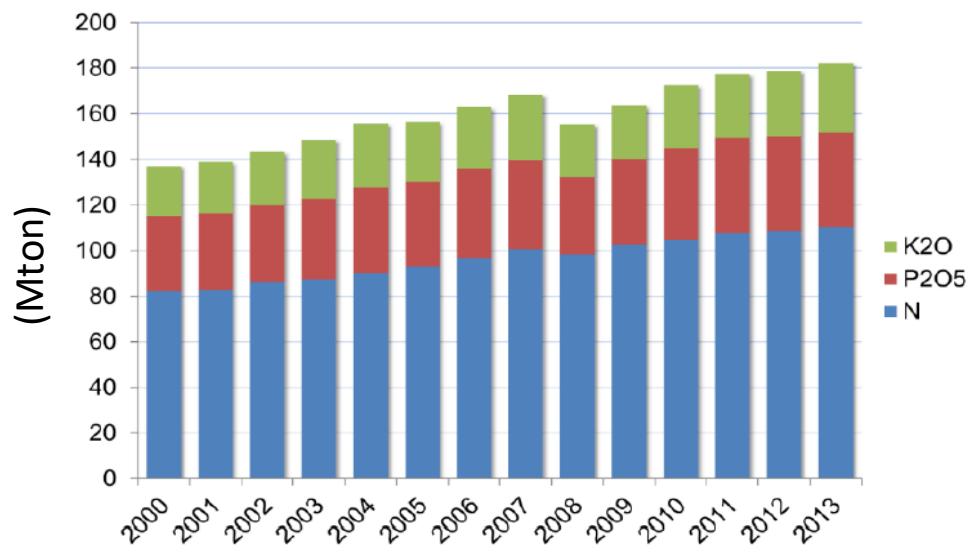
# Global trends



Source: United Nations, Department of Economic and Social Affairs, Population Division (2017).  
*World Population Prospects: The 2017 Revision*. New York: United Nations.

## World population

- Global population has been increasing
- It is expected to increase up to > 9 billion people by 2050 (and maybe > 11 billion people by 2100)
- Food production will have to increase by at least 60%

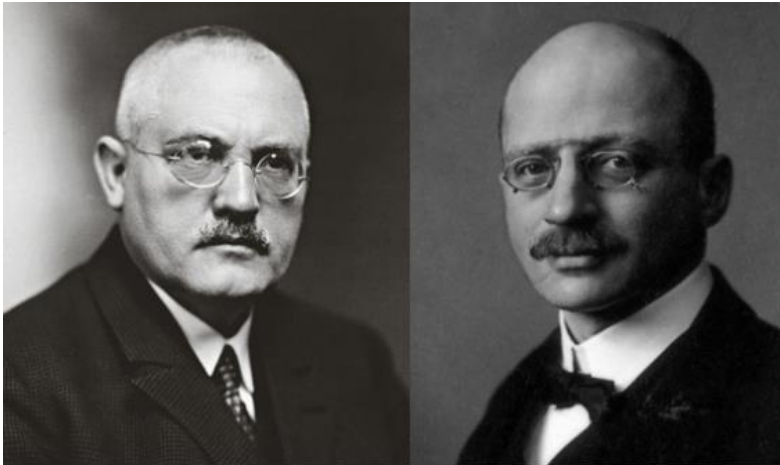


## Fertilizers

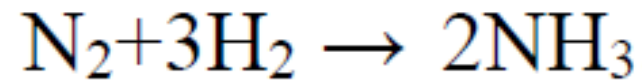
- In 2013-2014 the N fertilizers demand was of 110 Mton/y
- Fertilizers demand is expected to increase by 1.6% per year until 2021



# The Haber-Bosch process



Chemical N fixation



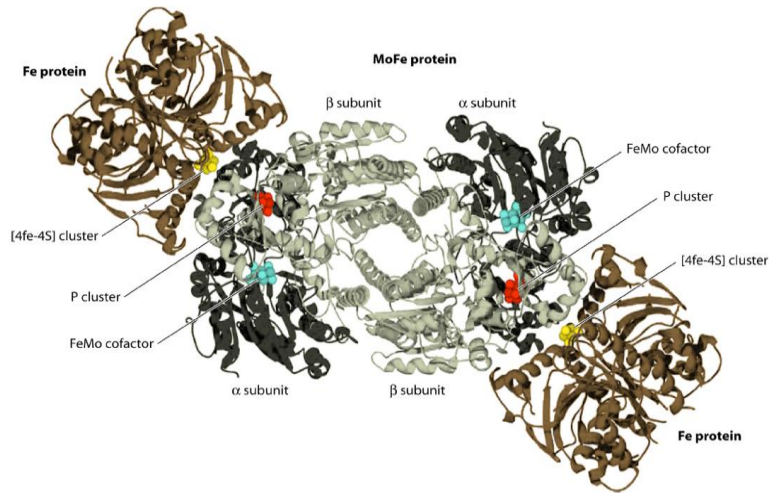
- High temperatures (700K) and pressures (100atm);
- 300 Mt of CO<sub>2</sub> emissions each year;
- Exploits 2% of global natural gas reserves;
- Has reached its maximum energy efficiency



**NOT SUSTAINABLE!**



# Can we find a greener process?

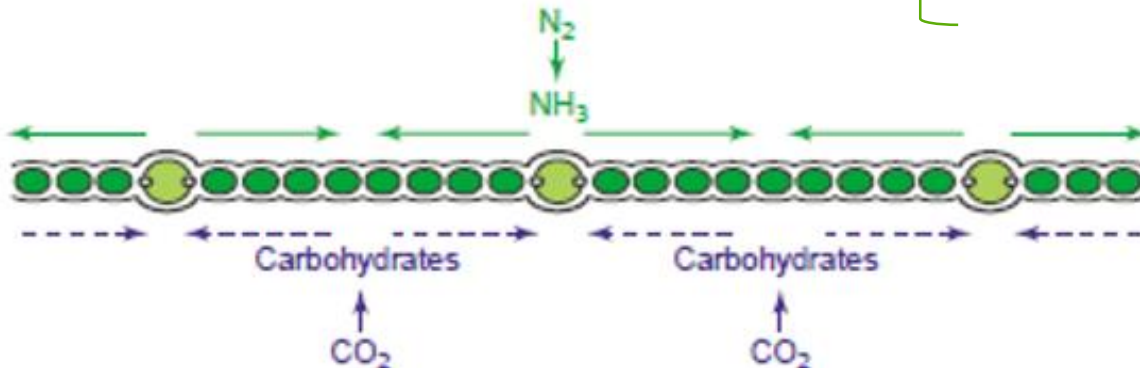


## Biological N fixation



## N-FIXING CYANOBACTERIA

- Photoautotrophs
- Largely distributed in aquatic and terrestrial ecosystems
- Could be of interest for industrial exploitation!



# Anabaena PCC 7122



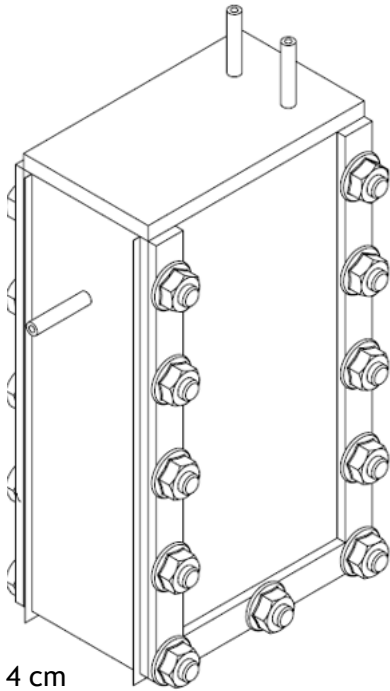
- Filamentous
- Heterocystous
- High specific growth rate ( $0.4 \text{ d}^{-1}$  in batch systems) under different light intensities



- Can it be cultivated in continuous photobioreactors?
- What fixed N productivity can be achieved?
- What are possible applications towards a sustainable agriculture?



# Continuous flat-panel PBR



## Experimental conditions

Cultivation medium: BG11<sub>0</sub> 2x  
Air-CO<sub>2</sub> (5% v/v)  
2.5 g L<sup>-1</sup> NaHCO<sub>3</sub>  
T = 24°C



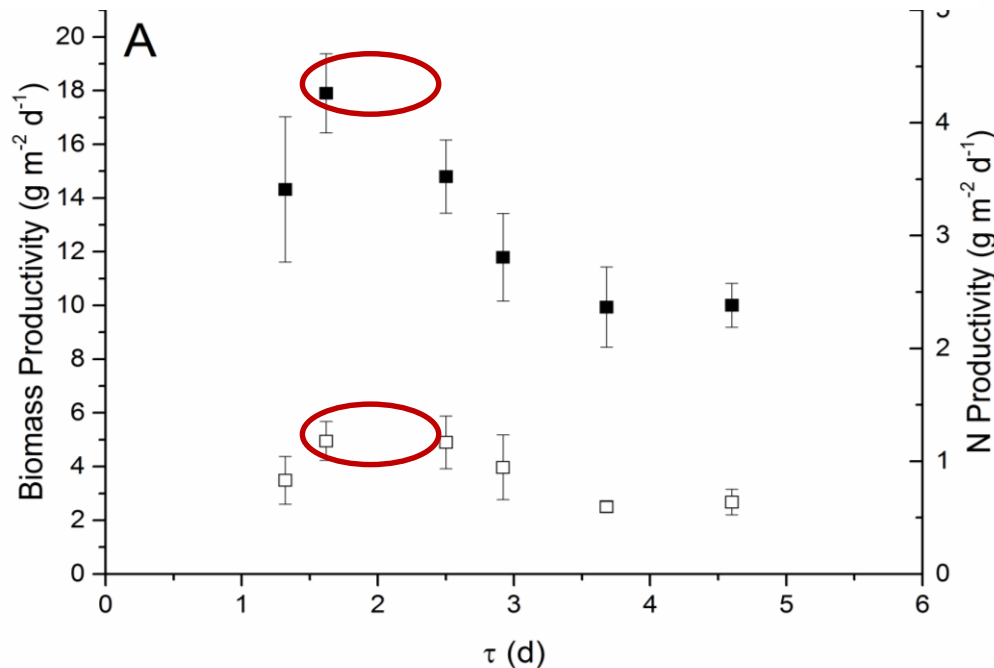
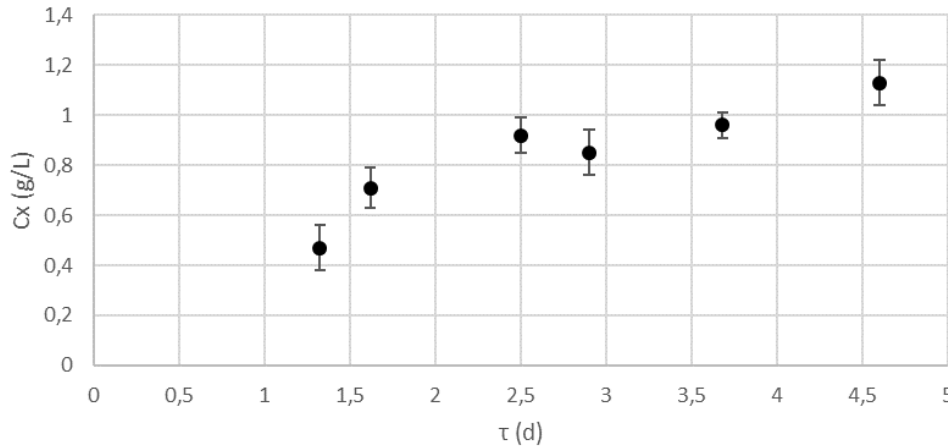
Effect of residence  
time and light  
intensity on biomass  
and N productivities



# Effect of residence time $\tau$

I:  $190 \mu\text{mol m}^{-2} \text{s}^{-1}$

$$\tau = V_R/Q$$



- N content in the biomass between  $5.73 \pm 0.82$  e  $7.88 \pm 0.85$  wt%, not influenced by  $\tau$

- Optimum value of  $\tau$  around **1.7 d**

- Maximum biomass productivity achieved:  $0.45 \text{ g L}^{-1} \text{ d}^{-1}$  /  **$18 \text{ g m}^{-2} \text{ d}^{-1}$**

- Maximum N productivity:  $30 \text{ mg L}^{-1} \text{ d}^{-1}$  /  **$1.2 \text{ g m}^{-2} \text{ d}^{-1}$**

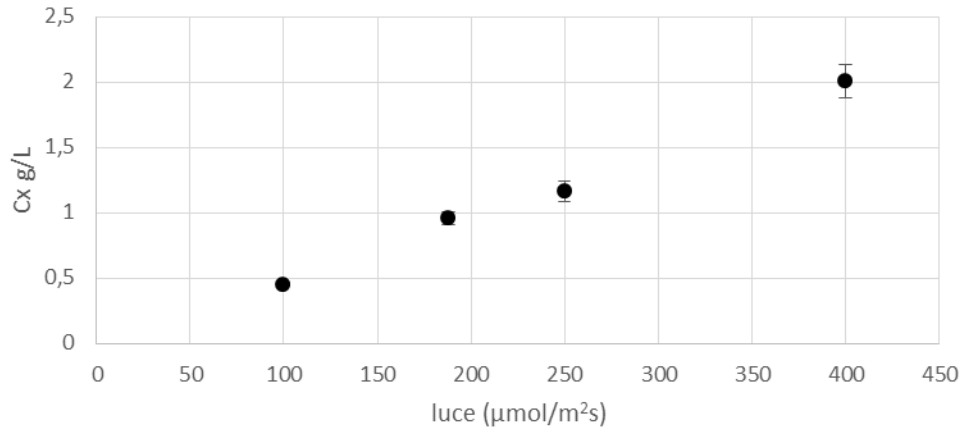


> 10x the global N fixing capability estimated in the ecosystem

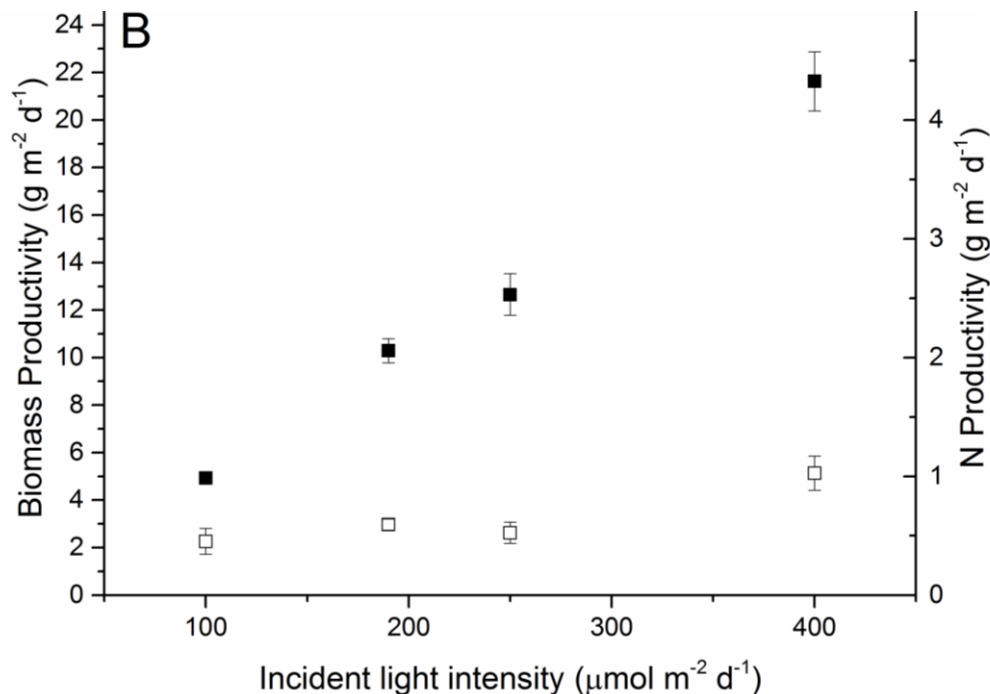


# Effect of incident light intensity

$\tau$ : 3.68 d



- No photosaturation/ photoinhibition effect up to  $400 \mu\text{mol m}^{-2} \text{s}^{-1}$
- Biomass productivity achieved:  $0.54 \text{ g L}^{-1} \text{ d}^{-1}$  /  $21 \text{ g m}^{-2} \text{ d}^{-1}$
- N wt% in the biomass appears negatively affected by the light intensity, but overall productivity increases

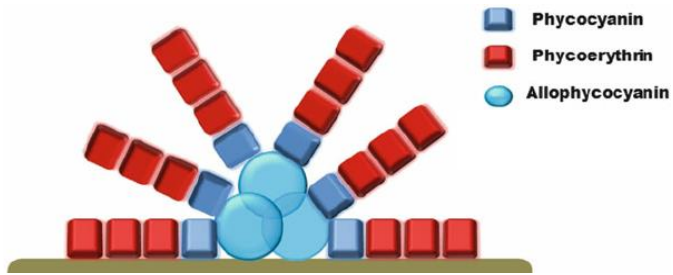


$I_{in}$ ( $\mu\text{mol m}^{-2} \text{s}^{-1}$ )	N wt%
100	$9.16 \pm 1.89$
190	$6.09 \pm 0.18$
250	$4.15 \pm 0.42$
400	$4.75 \pm 0.39$

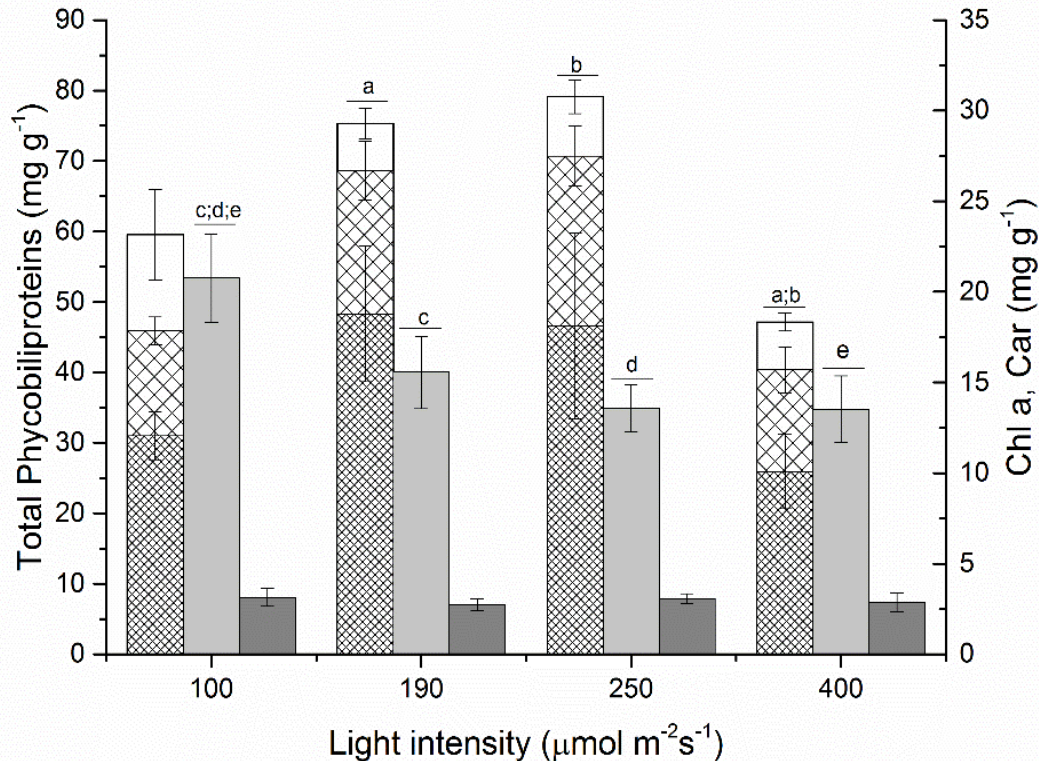




# Pigments (phycobiliproteins)



Light harvesting antennae: content is reduced when the light intensity increases, as a photoprotection system



High-value product  
(food/cosmetic  
industry):  
Productivity of about  
**1 g m<sup>-2</sup> d<sup>-1</sup>**



# Agricultural applications



## BIOFERTILIZERS



- Average N fertilization rate:  
160 kg N ha<sup>-1</sup> y<sup>-1</sup>
- Lab-scale experimental productivity achieved:  
4380 kg N ha<sup>-1</sup> y<sup>-1</sup> ✓

(Need to assess the actual efficiency)

## BIOSTIMULANTS

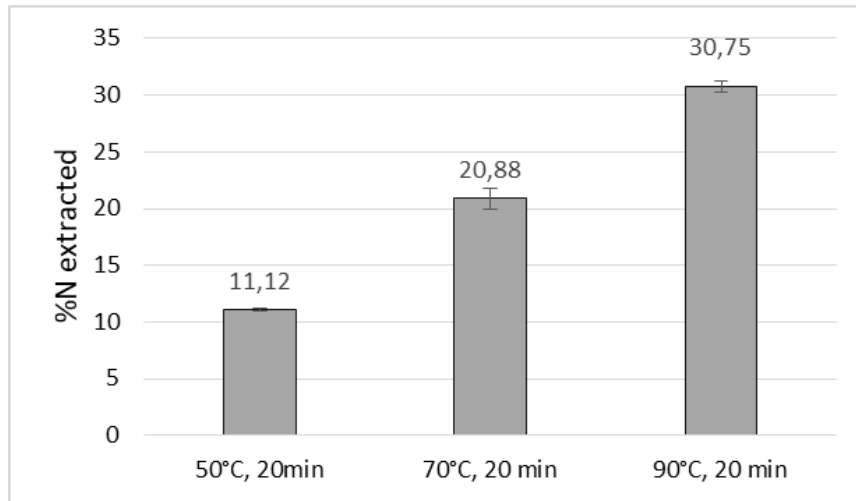


Different compounds of various origins and complex composition,  
characterized by some *hormon-like* activities, which increase crop productivity

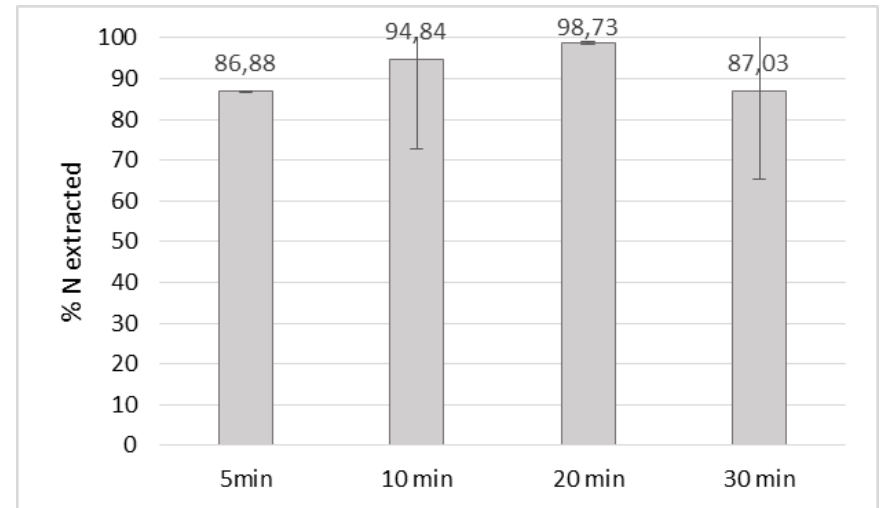
# Biostimulants

Three different pre-treatments to break the cell wall and extract compounds of possible interest

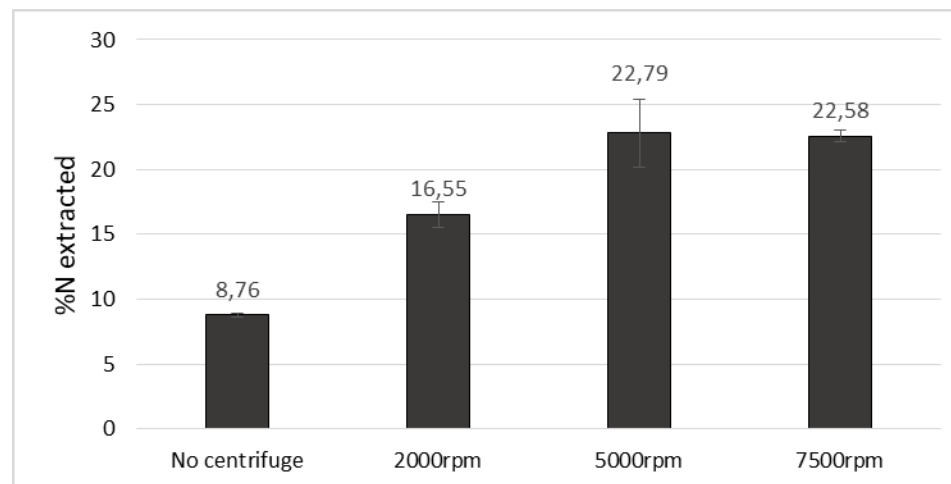
## Thermal



## Sonication

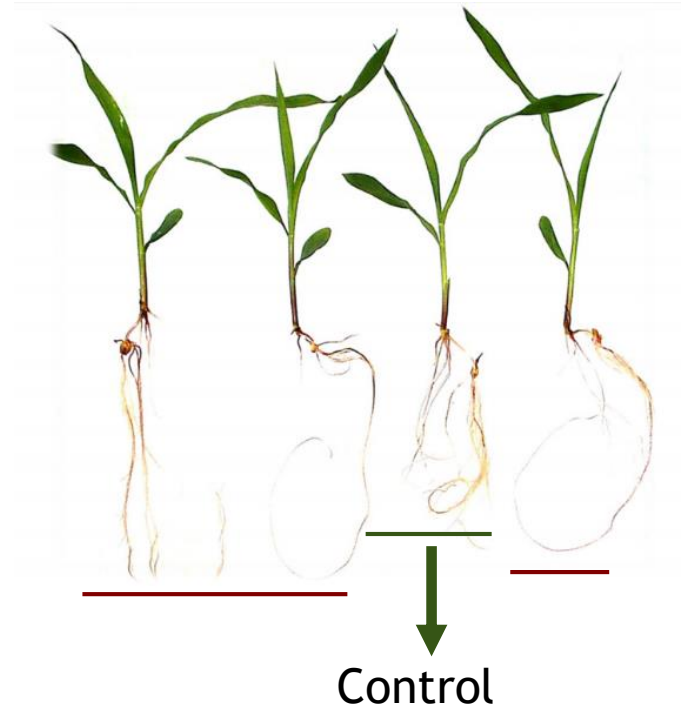


## Freeze/thaw + centrifuge



# Biostimulants

- Whole biomass: no *hormon-like* activity ✗
- Surnatant after freeze/thaw + centrifuge treatment: *auxin-* and *giberellin-like* activity ✓
- Surnatant after thermal treatment ?
- Surnatant after sonication treatment ?



# Conclusions

- *Anabaena* PCC 7122 was cultivated diazotrophically in continuous systems
- Nitrogen productivity achieved of  $12 \text{ kg N ha}^{-1} \text{ d}^{-1}$
- N content in the biomass is reduced under higher light intensity (phycobiliproteins)
- Potential for application in biofertilizers production
- Promising preliminary results for application as biostimulants

# Acknowledgments

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# Thank you for your attention

