# Innovative technologies to eliminate ammonia inhibition in anaerobic digestion in order to enhance methane production: Energy recovery from microalgae production

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## Abstract

The aim of this work is the study of the valorization of microalgae cultivated in anaerobic digestate with high ammonia content, towards energy production. Microalgae were grown in a photobioaeractor under varying conditions, aiming to the determination of the effect of substrate composition and nitrogen concentration on the biomass composition and the corresponding energy production potential. *Chlorella sorokiniana* species were cultivated in a 20 L photobioreactor using standard BG11 nutrient substrate or digestate from an anaerobic biogas unit, with varying ammonia nitrogen concentrations. The produced biomass was then subjected to analysis of the composition for the determination of its content in fats, hydrocarbons, and proteins. The cultivated microalgae biomass presented high biochemical methane production potential, reaching up to 350 mL $CH_4$/g VS. The potential can be enhanced by the prehydrolysis of cells: pretreatment at 40°C for a contact time of 4 to 10 hours increased the methane production potential to 455 mL $CH_4$/g VS.

## Experimental Methods

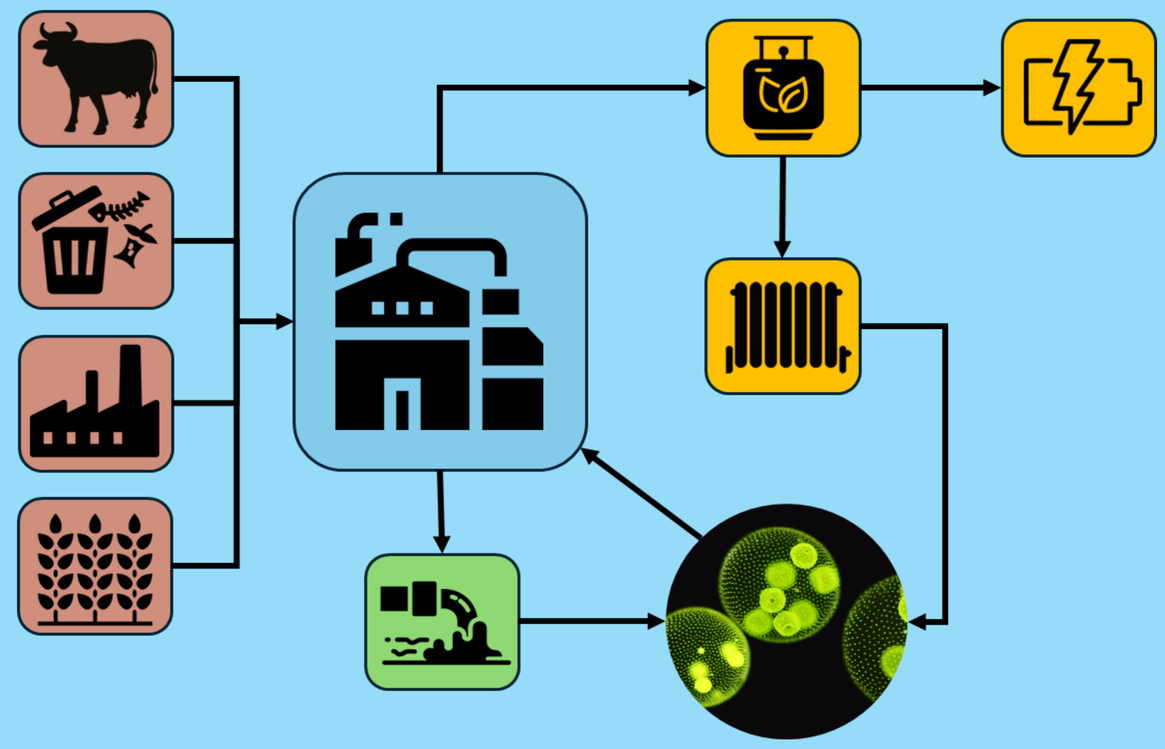
**Experimental Setup**

- Semi-pilot scale, horizontal, tubular photo-bioreactor with operating volume of 20 L
- Ultrafiltration membrane for the collection of the cultivated microalgae
- Thermal pretreatment at 40°C and 90°C

**Analytical Methods**

- Determination of intracellular proteins with the micro BCA method
- Determination of intracellular carbohydrates with ultraviolet-visible spectroscopy
- Determination of intracellular lipids with ultraviolet-visible spectroscopy
- Determination of a,b chlorophyl and carotenoids with ultraviolet-visible spectroscopy
- Determination of total solids, volatile solids and dissolved COD
- Determination of dissolved proteins and carbohydrates
- Determination of biochemical methane production potential

## Background

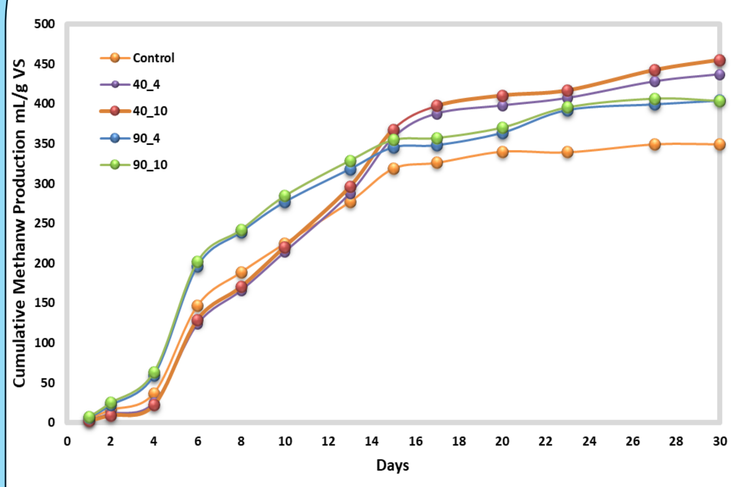


The need to address environmental problems arising from the increasing use of fossil fuels to meet modern energy demand and the induced $CO_2$ accumulation in the atmosphere requires the use of innovative methods of clean energy production and waste management. The production of bioenergy utilizing microalgae biomass is promising as a potential alternative energy source. Microalgae can be cultivated alongside various waste treatment systems, improving their efficiency and the bioenergy produced fulfills all the criteria of a renewable energy source. A potential method of utilizing the microalgae biomass can be through the anaerobic digestion process for the production of biogas.

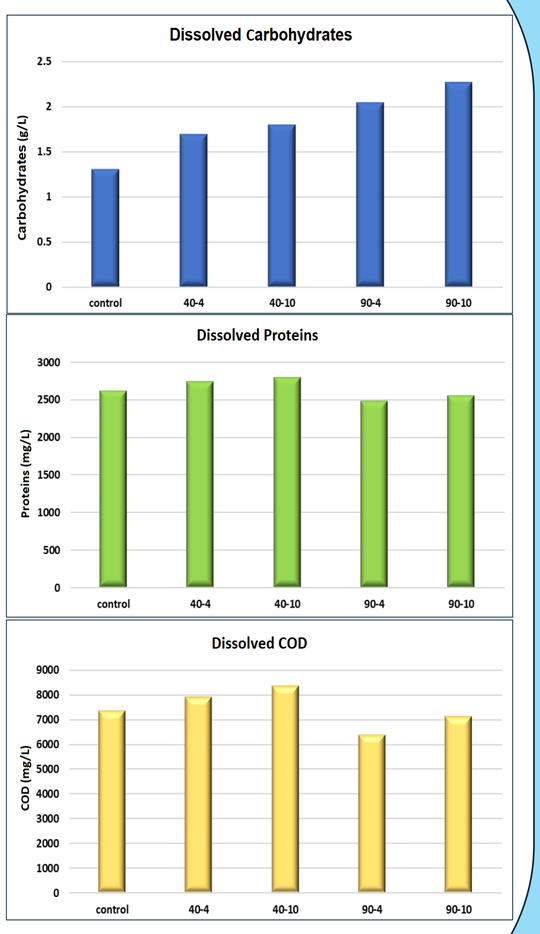
## Results and Discussion

Photo-bioreactor used to cultivate microalgae and ultrafiltration membrane tank

### Biogas production from the anaerobic digestion of microalgae

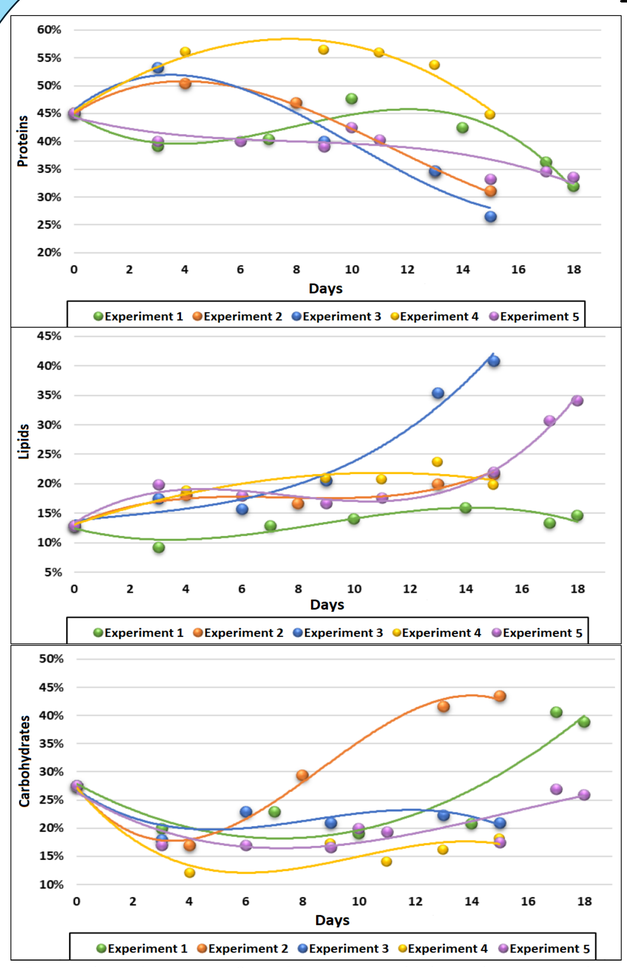



Biochemical methane production potential of microalgae: biomass with no pretreatment (control), biomass with thermal pretreatment at 90°C for 4 hours (90_4) and at 90°C for 10 hours (90_10), biomass with thermal pretreatment at 40°C for 4 hours (40_4) and at 40°C for 10 hours (40_10)

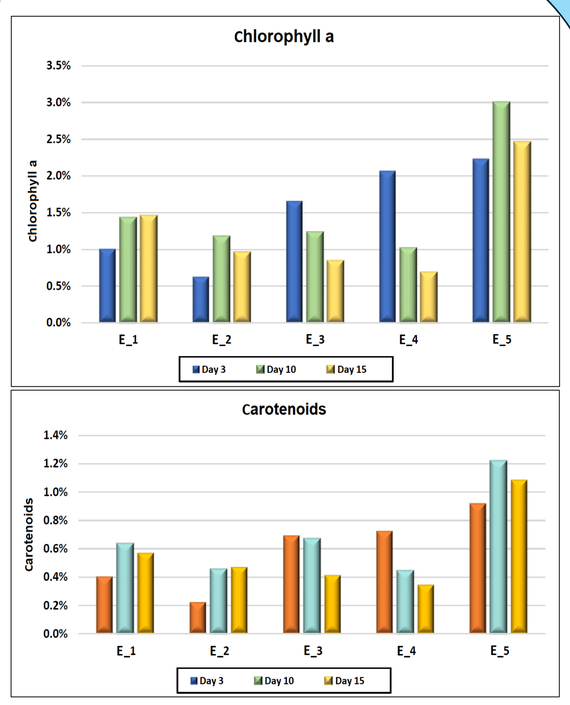



Dissolved carbohydrates(a)proteins(b)and COD(c)in biomass after pretreatment: no pretreatment (control), thermal pretreatment at 90°C for 4 h(90_4) and 10 h(90_10), thermal pretreatment at 40°C for 4 h(40_4) and 10 h(40_10)

### Effect of cultivation conditions and substrate on the biomass composition




Dynamic change of cell composition in proteins (a), lipids (b) and carbohydrates (c) of photo-bioreactor batch operation tests




Composition of cells in chlorophyll a and carotenoids in three different days during the photo-bioreactor batch operation tests

## Conclusions

- *C. Sorokiniana* was cultivated in the effluent from an anaerobic digestion unit treating livestock production residues, with ammonia content of about 4-5 g/L.
- Microalgae can be used for biogas production: biochemical methane potential reached up to 350 mL/g VS.
- Biomass pretreatment at 40 °C enhances methane production that can reach up to 455 mL $CH_4$/g VS and is considered the most effective technique based on energy requirements.
- Regardless of the pretreatment technique, the majority of biogas, 80-90%, is produced within 15 days.
- The composition of microalgae species depends on the availability of nutrients.
- At low nitrogen and/or phosphorus conditions, the intracellular composition is strongly altered and lipid and/or carbohydrate overaccumulation is observed. In the present work carbohydrates accumulated up to 43.5% and lipids up to 40.8%.
- In the continuous operation of the waste treatment-biomass producing system, the composition of biomass was: carbohydrates 28.5%±0.45%, lipids 20.8%±0.31% and proteins 50.5%±1.96%.
- During the cultivation of the strain at high concentration substrate (≥5%), the percentage of chlorophyll a increases, with the highest value being 3%.

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