

# EVALUATION OF THE ANTIMICROBIAL ACTIVITY AND DIGESTIBILITY OF ALGAE CULTIVATED ON SIDESTREAMS

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## Introduction

Food and feed are some of the potential application areas for algae cultivated on sidestreams. Apart from safety and legal requirements, the digestibility of the produced algae biomass should be assessed to evaluate this potential. Moreover, research literature suggests that algae (fractions) can have antimicrobial activities that could be of interest for many applications.

## Aim

The goal of this work was twofold: (i) to study the digestibility of algae cultivated on different sidestreams, and (ii) to perform a first evaluation of the antibacterial activity and antifungal activity of algae grown on sidestreams.

## Conclusions

- The rather high N (57%-69%) and organic matter (51-64%) solubility after digestion suggested that most of the proteins and organic matter are accessible for enzymatic degradation in the gut.
- Several ethanol extracts showed antimicrobial activity. Further research is now needed to confirm this activity and to identify the antimicrobial compounds.

## Materials & methods

The table below details the algae and side streams used for algae cultivation.

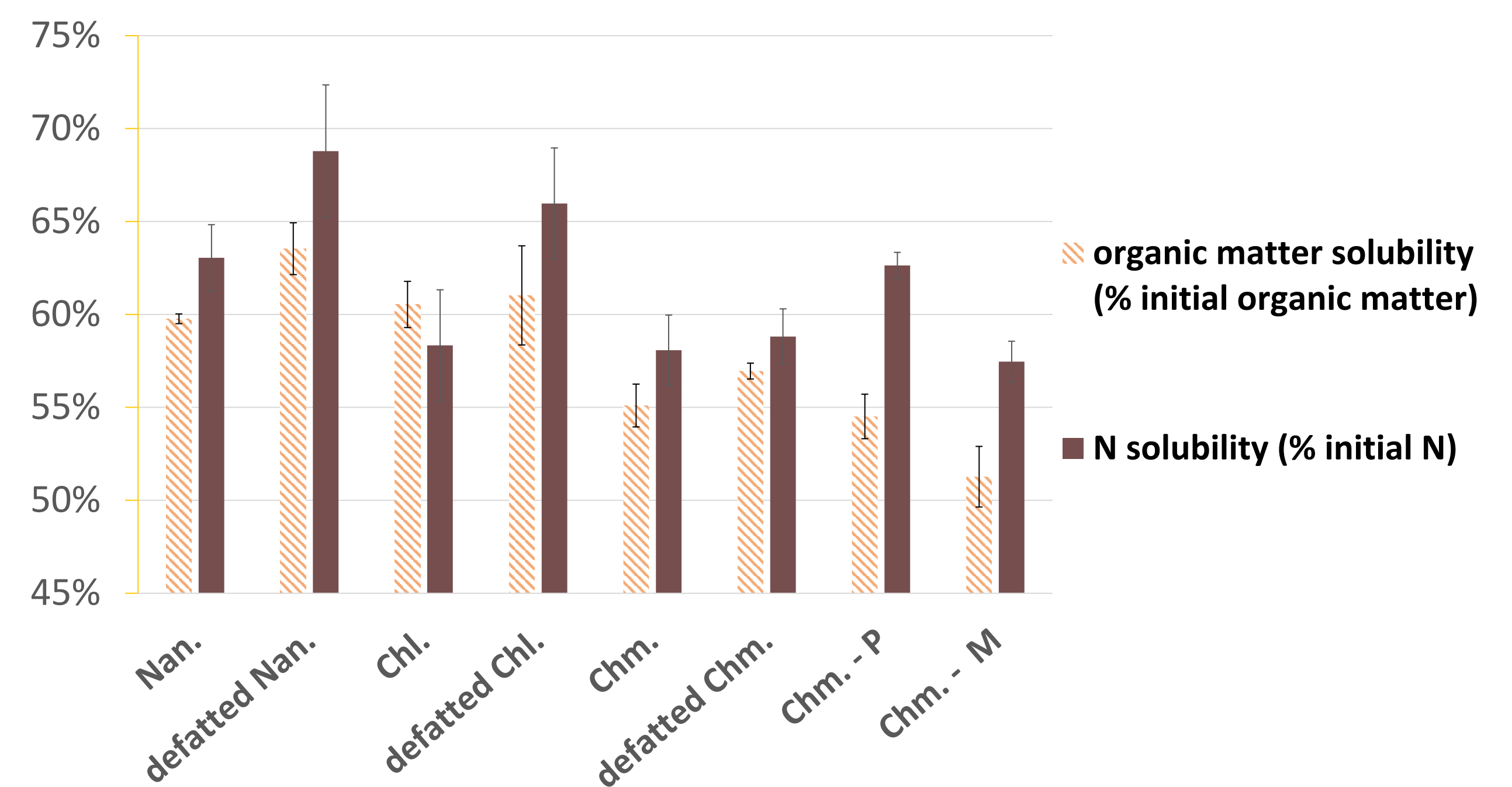
Algae	Cultivation	Downstream processing	Code
<i>N. gaditana</i>	Using permeate obtained by membrane filtration of an open pond culture	Cell disruption	Nan
		Cell disruption and defatting	Defatted nan
		Oil	Nan oil
<i>Chlorella sp.</i>	Using CO <sub>2</sub> from biogas combustion	Cell disruption	Chl
		Cell disruption and defatting	Defatted Chl
		Oil	Chl oil
<i>Chloromonas</i>	Using permeate obtained by membrane filtration of an open pond culture	Cell disruption	Chm
		Cell disruption and defatting	Defatted Chm
		Oil	Chm oil
	Using permeate obtained by membrane filtration of an open pond culture	Cell disruption	Chm - P
	Using regular growth medium and in the same time period as Chm-P	Cell disruption	Chm - M
Scenedesmus	Using treated digestate	Cell disruption	Scen
Mixed algae culture	Using permeate obtained by membrane filtration of an open pond culture	Cell disruption	Mixed

- Algae digestion** was simulated according to the INFOGEST protocol (Brodkorb 2019) after which the soluble fraction was separated by centrifugation. The organic matter and N solubility were evaluated to get a first indication of the amount of organic matter and protein that is available to enzymatic degradation in the gut.
- Ethanol extracts were prepared from algae to evaluate the **antibacterial activities**. The growth curve of *E. coli* and *S. aureus* with or without the presence of an ethanol extract, solubilized in DMSO, was established by turbidity measurements.
- To evaluate the **antifungal activity**, the same extracts were applied to tomato leaves and after infection with spores of *Botrytis cinerea* and an incubation period, the infected surface area was measured.

## Results

### Digestibility

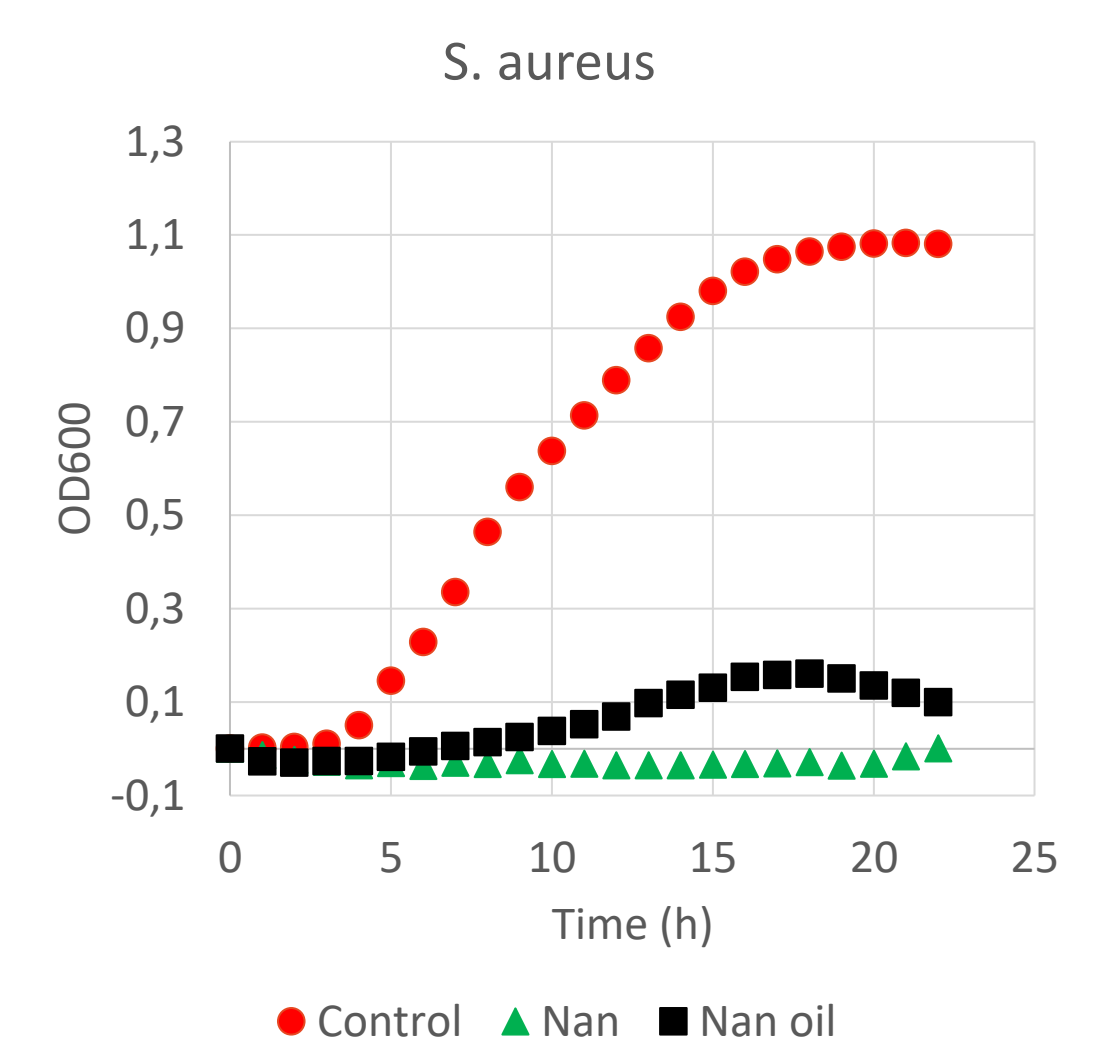
The solubility after *in vitro* digestion gives a first idea about algae's digestibility as the biomass that is insoluble after digestion, can not be taken up in the small intestine.



The majority of the organic matter was soluble after digestion with solubility levels ranging between 51 and 64%. Similarly, most N was soluble and N solubility levels after digestion between 57% and 69% were noted. Slightly higher organic matter and N solubility levels were observed for *Chloromonas* grown on permeate than that grown on a regular medium. Note that all samples were disrupted before digestion testing.

### Screening of antibacterial activity

	<i>E. coli</i>		<i>S. aureus</i>	
	% OD reduction at 8 h	% OD reduction at 20 h	% OD reduction at 8 h	% OD reduction at 20 h
Nan	15	47	100	100
Defatted nan	-8	12	69	19
Nan oil	-13	12	97	88
Chl	67	100	87	93
Defatted Chl	72	70	69	74
Chl oil	-1	60	85	86
Chm	-2	36	83	87
Defatted Chm	-59	34	67	74
Chm oil	44	60	100	100
Chm - P	-15	94	87	96
Chm - M	-55	74	86	100
Mixed	-25	93	100	100



Some ethanol extracts of the algae samples at 1 g/L (indicated in green in the above table) had an impact on the growth of the bacteria by slowing down or preventing growth, resulting in a lower bacterial load after 24h contact.

### Screening of antifungal activity



The figure above illustrates the different *B. cinerea* infection zones of leaves without algae extracts (blank) and those with extracts. The *Nannochloropsis* extract (*Nan.*) showed no antifungal activity, while antifungal activity was observed for the following algae extracts: Nan oil, Chl oil, Chm -M and Mixed.

