



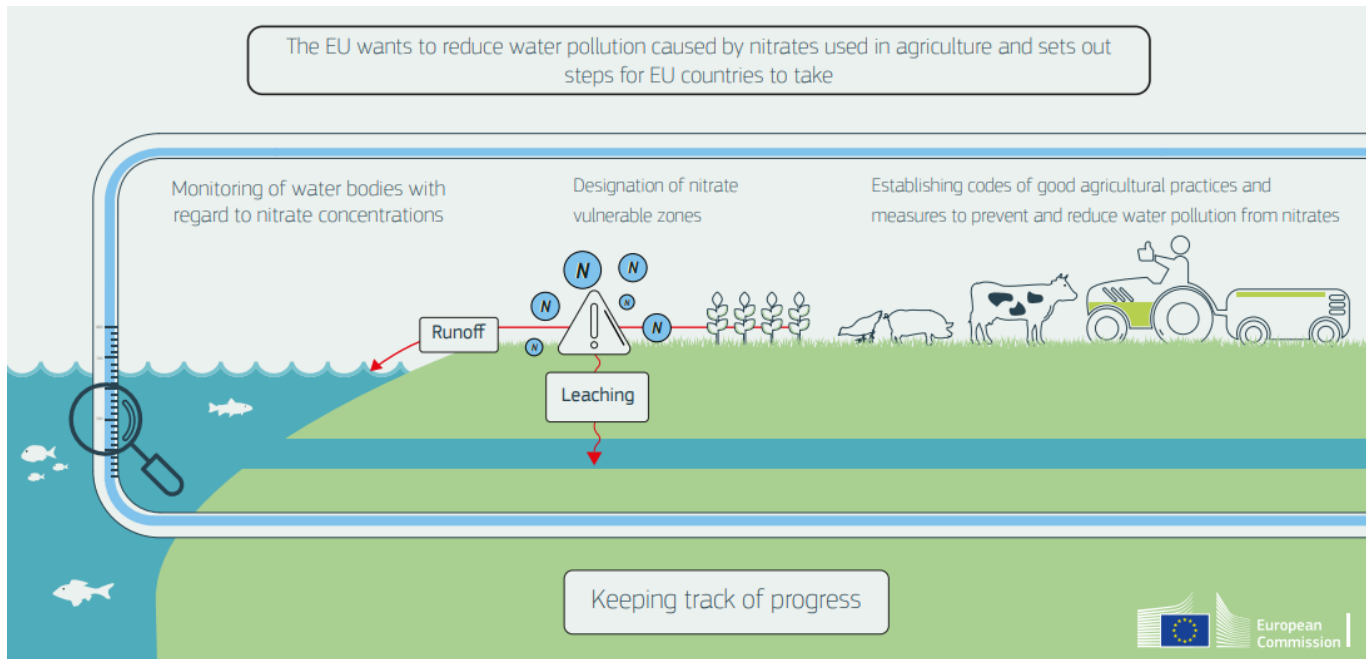
The ALG-AD Project: *Creating value from waste nutrients by integrating algal and anaerobic digestion technology*

Fleuriane Fernandes

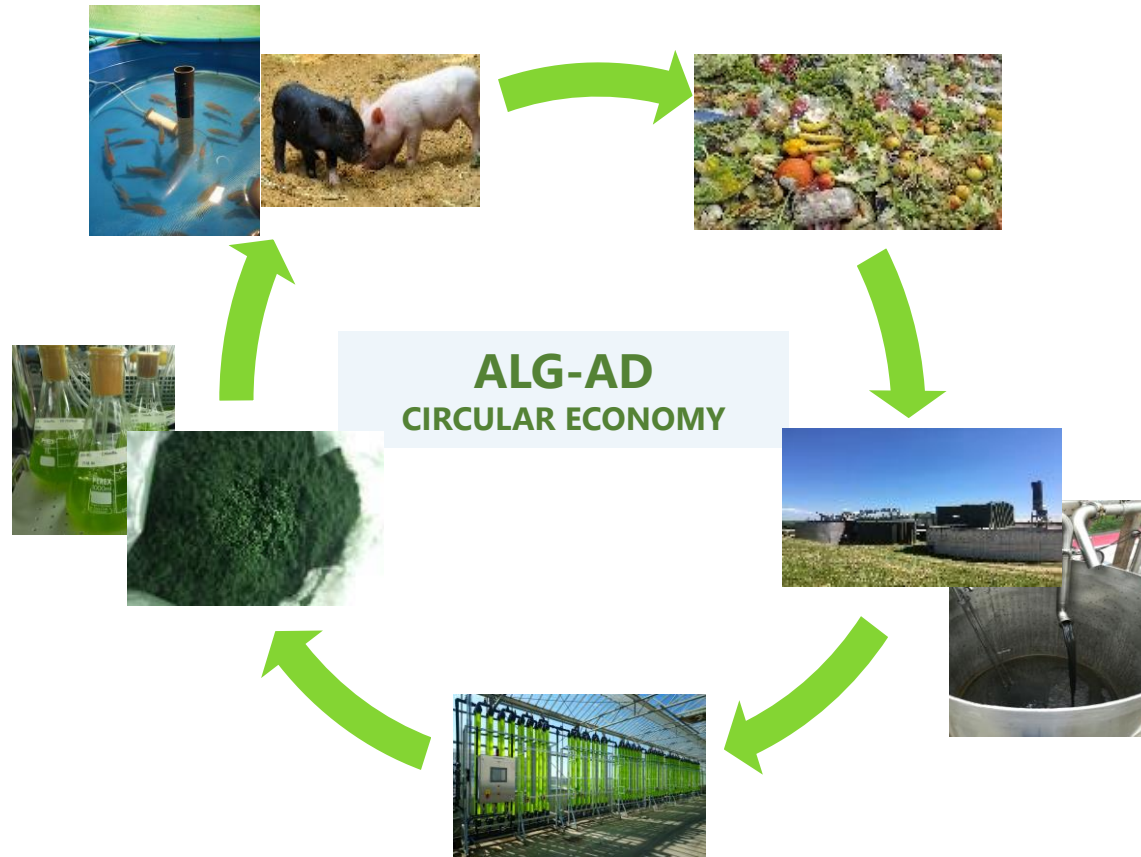
29th of March 2022



The ALG-AD project Rationale: The European Nitrate Directive



The ALG-AD Project: the circular economy concept



ALG-AD combines algal and AD technology to reduce excess nutrients in liquid digestate

ALG-AD converts nutrients to create algal biomass for sustainable animal feeds

Three 4 tonne pilot facilities to implement and test the technology in real life conditions

11 partners from North West Europe (NWE) including the UK, France, Belgium and Germany

The biomass produced has been used to conduct piglet and fish feeding trials

Information collated has enabled the development of Decision Support Tools

DIGESTATE TREATMENT FOR MICROALGAE CULTIVATION



Interreg
North-West Europe
ALG-AD

ALG-AD-Project no. NWE 520

Public Output

Output WP1A1.1:

Best Practices for the treatment and preparation of nutrient rich digestate for algal cultivation

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Digestate

- Dark Colour
- High dry weight – Particles
- N & P concentration -> to be tailored to microalgal needs



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Valourising nutrient-rich digestate: Dilution, settlement and membrane filtration processing for optimisation as a waste-based media for microalgal cultivation

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ABSTRACT

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Keywords:
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Settlement and dilution
Microalgae
Chlorella vulgaris
Pilot-scale

Digestate produced from the anaerobic digestion of food and farm waste is primarily returned to land as a biofertiliser for crops, with its potential to generate value through alternative processing methods at present under explored. In this work, valorisation of a digestate resulting from the treatment of kitchen and food waste was investigated, using dilution, settlement and membrane processing technology. Processed digestate was subsequently tested as a nutrient source for the cultivation of *Chlorella vulgaris*, up to pilot-scale (800L). Dilution of digestate down to 2.5% increased settlement rate and induced release of valuable compounds for efficient usage such as nitrogen and phosphorus. Settlement, as a partial processing of digestate offered a physical separation of liquid and solid fractions at a low cost. Membrane filtration demonstrated efficient separation of nutrients, with micro-filtration recovering 92.38% of phosphorus and the combination of micro-filtration, ultra-filtration, and nano-filtration recovering a total of 94.33% of nitrogen from digestate. Nano-filtered and micro-filtered digestates at low concentrations were suitable substrates to support growth of *Chlorella vulgaris*. At pilot-scale, the microalgae grew successfully for 28 days with a maximum growth rate of 0.62 day⁻¹ and dry weight of 0.86 g L⁻¹. Decline in culture growth beyond 28 days was presumably linked to ammonium and heavy metal accumulation in the cultivation medium. Processed digestate provided a suitable nutrient source for successful microalgal cultivation at pilot-scale, evidencing potential to convert excess nutrients into biomass, generating value from excess digestate and providing additional markets to the anaerobic digestion sector.

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1. Introduction

Anaerobic digestion (AD) is commonly used in Europe for the treatment of food and farm waste. The AD process is a biological mechanism during which bacterial and archaeal communities convert carbon-rich organic waste into biogas, primarily methane and carbon dioxide (Doyle and Kumar, 2005). Another by-product of the AD process is a nutrient-rich digestate (NRD). NRD is rich in carbon, nitrogen (N), phosphorus (P) and other macro and micronutrients (Papadimitriou et al., 2006; Tomlinson et al., 2017). NRD is primarily used as organic fertilizer and is directly applied onto farmland (Fuchs and Drogg, 2013). However, the use of digestate as a soil fertilizer increases the risk of nutrient runoff and penetration of groundwater resources, leading to soil and water eutrophication (Gullayn et al., 2019). Consequently, Nitrate Vulnerable Zones (NVZs) have been designated under the European Nitrate Directive 91/676/EEC that limits the annual load of nitrogen applied onto farmland. NVZs are on the increase across North West Europe, resulting in the accumulation of approximately 10 million tons of excess digestate (Fuchs and Drogg, 2013). Alternatives to farmland spreading have been investigated, such as using solid digestate for energy production or conversion into added-value products (char or activated carbon) (Moulin et al., 2015), however valorisation of digestate has been underexplored and solutions have yet to be firmly established to create value from this excess NRD. The present study focused on mechanical and biological treatment of digestate to increase its value and marketability. The partial processing of digestate was investigated first, by establishing methods for the separation of liquid and solid fractions of digestate using simple low-cost techniques. This approach

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Membrane filtration was efficient at separating liquid and solid fractions of NRD and at recovering nearly 95% of both N and P

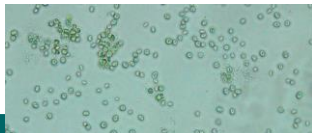
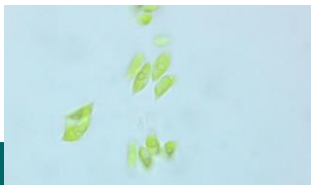
ALG-AD CULTIVATION PILOTS



Langage AD, Plymouth



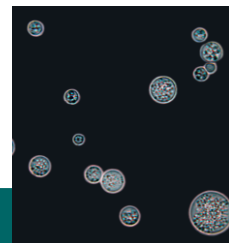
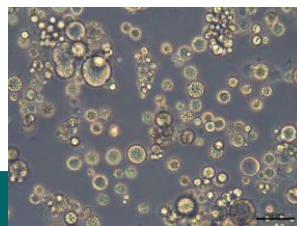
5T photobioreactor
Chlorella sp. and *Scenedesmus sp.*
2.5% of micro-filtered digestate (origin: kitchen waste)



Cooperl, Lamballe



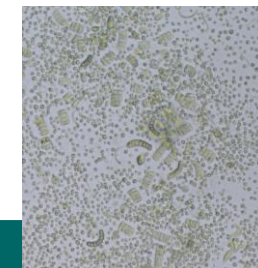
2T photobioreactor
Auriantochitrium mangrovei
80% of centrifuged digestate (origin: pig manure)



Innolab, Ghent



2T photobioreactor
Consortium of *Chlorella sp.* and *Desmodesmus sp.*
5% of filtered digestate (origin: plant and food waste)



ALG-AD CULTIVATION PILOTS – Biomass production



2.5% digestate (80 mg/L NH₄⁺)

7 days of cultivation

10 g/L glucose in mixo phase

14 g/L final biomass

20 mg/L/day N uptake

2.5% digestate (70 mg/L NH₄⁺)

2 days of cultivation

20 g/L glucose, 2 g/L yeast extract, 2 g/L peptone

4 g/L final biomass

35 mg/L/day N uptake

2.5% digestate (50 mg/L NH₄⁺)

7 days of cultivation

No external nutrients

1.7 g/L final biomass

20 mg/L/day N uptake



25 kg of concentrated biomass (10g/L) produced by pilots



Towards a circular economy: A novel microalgal two-step growth approach to treat excess nutrients from digestate and to produce biomass for animal feed

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HIGHLIGHTS

- A new two-phase microalgal growth process was developed.
- Microalgal cultures were able to grow on industrial waste and bioremediate nutrients.
- High quality microalgal biomass was obtained suitable for use as animal feed.

GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:
Circular economy
Microalgae
Digestate
Biomass production

ABSTRACT

Implementing a circular economy aimed at reusing resources is becoming increasingly important for industry. Microalgae fit within a circular economy by being able to bioremediate nutrient waste and as a source of biomass for several commercial applications. Here, we report a novel validation of a circular economy concept using microalgae as a nutrient industrial waste with a new two-phase process. During the first phase biomass was grown heterotrophically, biomass was then concentrated using membrane technology for the second phase where microalgal conditions were applied to boost growth further. Microalgal cultures were able to grow (12.8 g/L) on industrial wastewater (Digestate > 124 mg/L NH₄⁺) from an anaerobic digestion substrate (digestate), obtaining high quality microalgal biomass (>48% protein content) suitable for use as animal feed, closing the circular economy loop for industrial applications.

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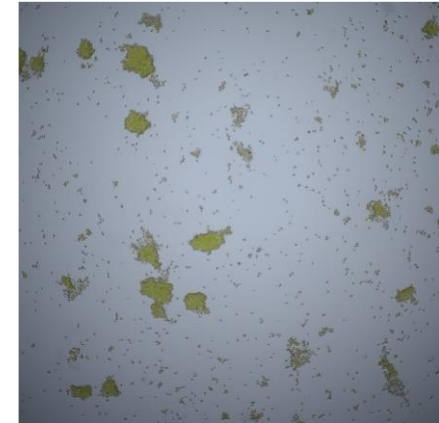
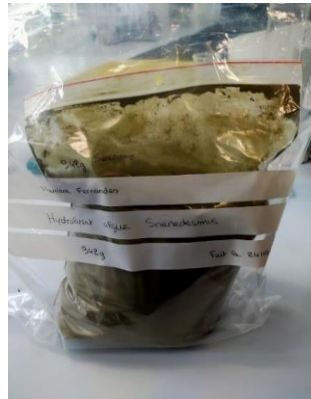
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BIOMASS PROCESSING - Hydrolysis



Specific hydrolysis process : enzymatic cocktails tailored to the different types of biomass

Breaking down of proteins and other molecules into peptides and amino acids and other metabolites of interest for feed trials



Cheaper than extraction methods

Better incorporation into feed formulation and better absorption by animals



Seabass Trials

- *Auranthiochytrium mangrovei* used to replace 15% of a standard feed for Seabass Juvenile and larvae
- Objective : Increase DHA supply and DHA/EPA ratio with microalgae biomass inclusion

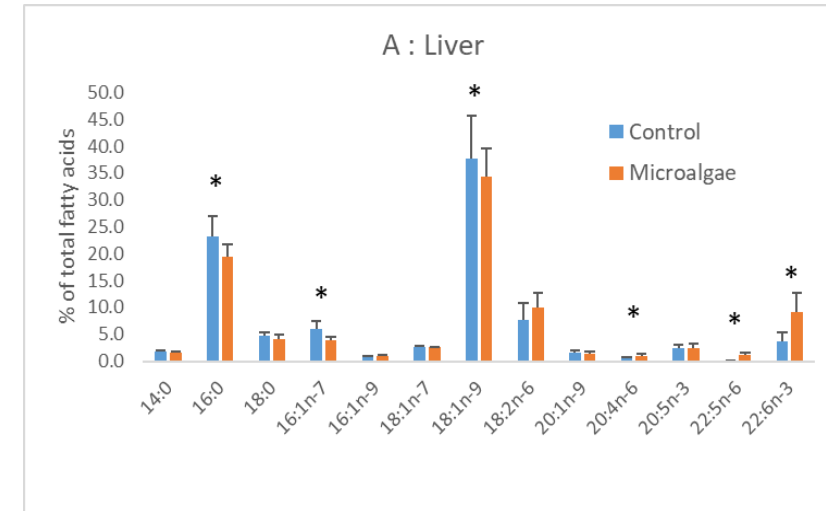
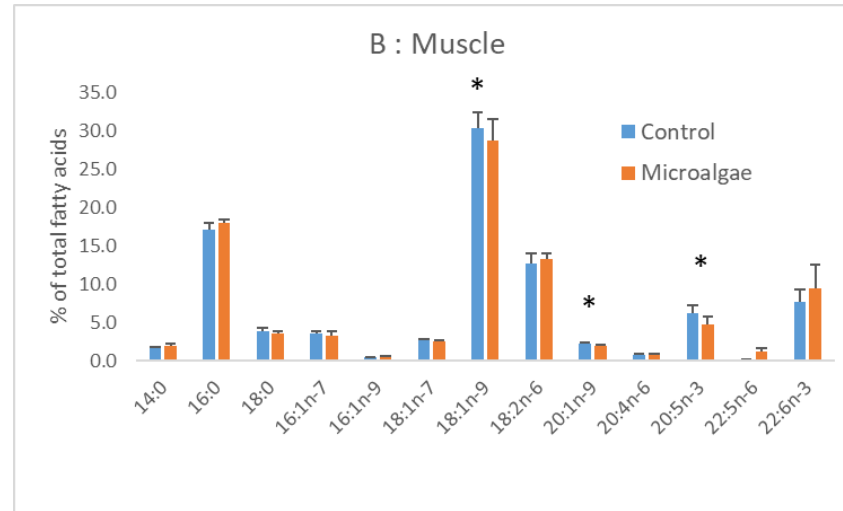
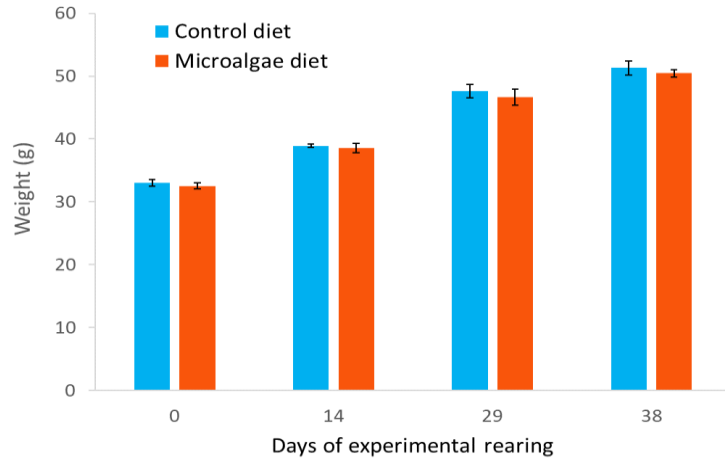
Tilapia Trials

- *Scenedesmus obliquus* used to replace 10% of a standard feed for Tilapia juveniles
- Objective : decrease reliance on fish meal proteins by replacing them with SO as a high source of proteins

ALG-AD FEED



SEABASS TRIAL MAIN RESULTS



Comparable growth of the fish

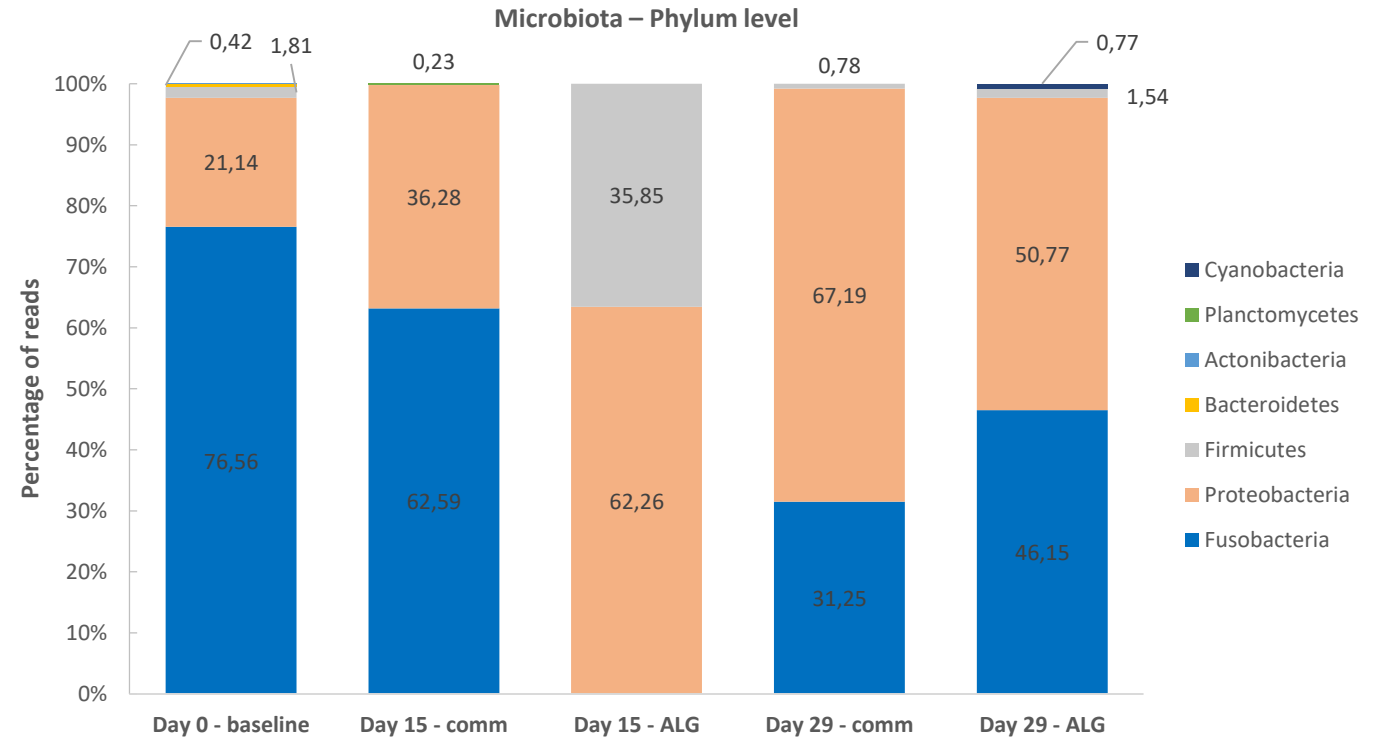
DHA increased in liver and in muscle -> added value for aquaculture sector



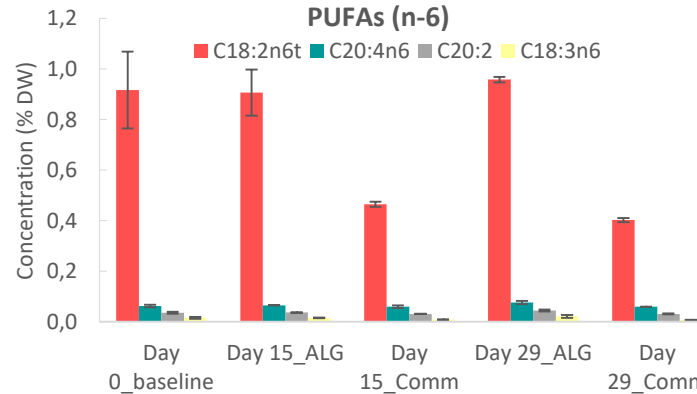
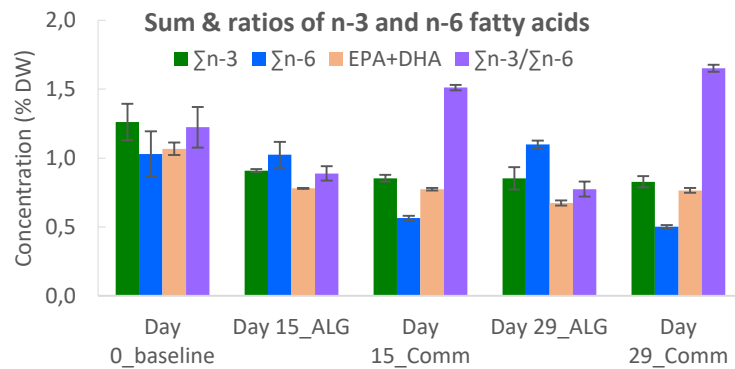
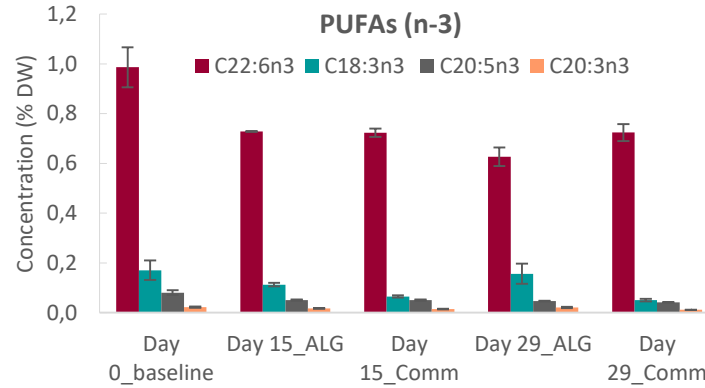
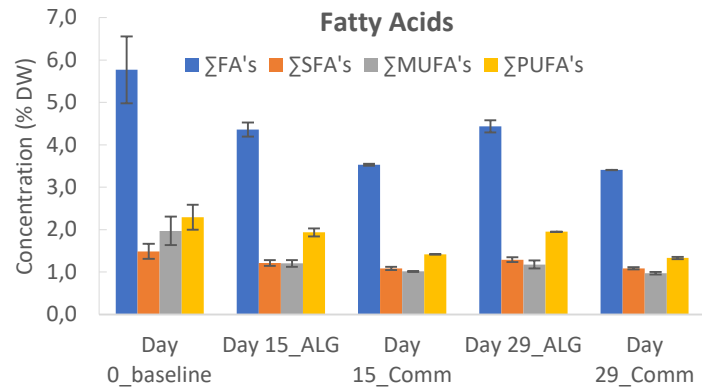
TILAPIA TRIAL MAIN RESULTS



Higher microbial diversity -> better fish health and immunity



TILAPIA TRIAL MAIN RESULTS



Twenty-two FAs found

Total fatty acids and PUFAs were significantly **higher** for the **ALG-AD feed**.

Fatty acid content of fish directly influenced by the **fatty acid composition of the diet** : **Large amounts of alpha-linolenic acid** in *Scenedesmus obliquus* -> metabolised in different lipids

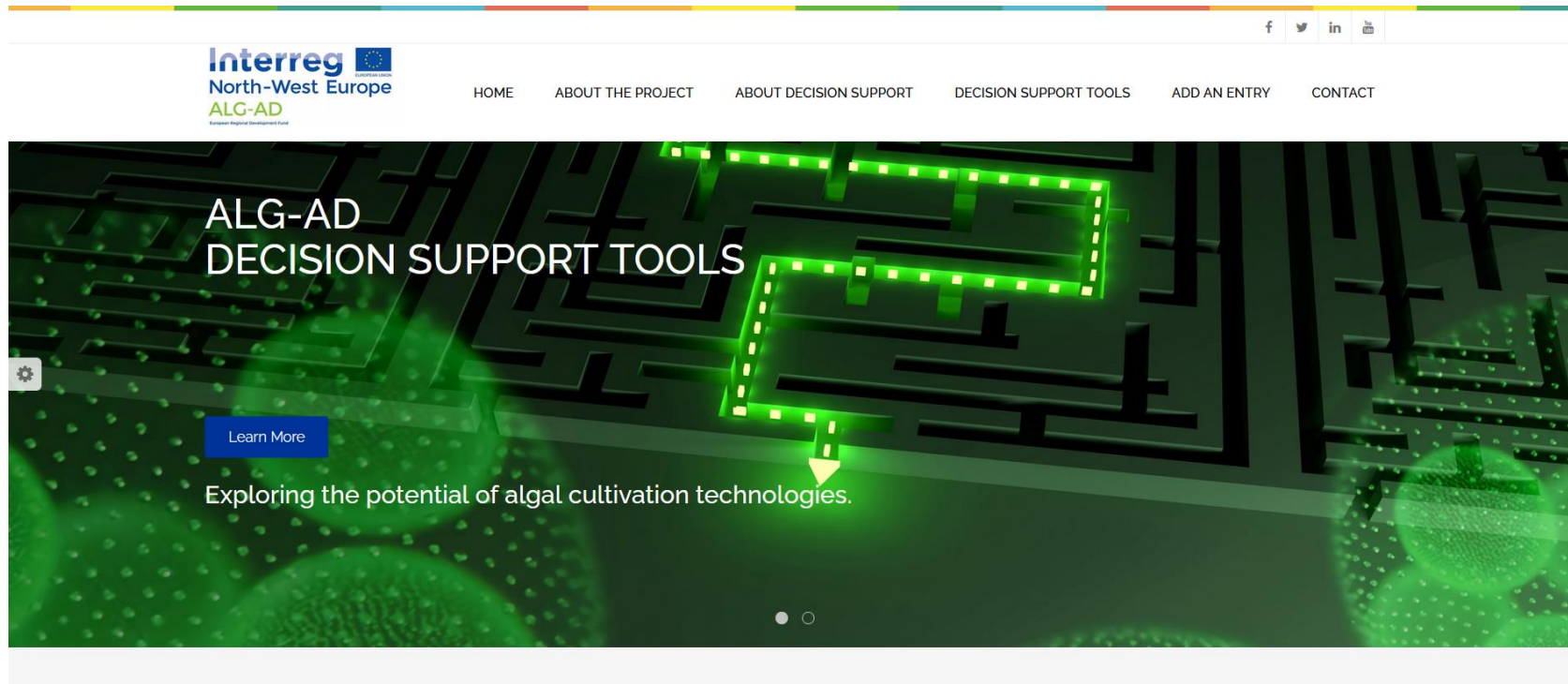
ω-3/ω-6 ratio of nearly 1 -> associated with lower risks of developing allergies, inflammatory and cardio-vascular diseases

Improved flesh quality and consequent added-value for the aquaculture sector

DECISION SUPPORT TOOL



Inform and guide technology developers, businesses, policy makers and researchers.



The tools can provide supporting information that is unique to your location and circumstances. They are flexible and adaptive to enable you to explore various process scenarios.

Release in the next few months

PUBLICLY AVAILABLE OUTPUTS AND DELIVERABLES



Interreg North-West Europe
ALG-AD
Greenhouse Gas Emission Fund

THEMATIC PRIORITY
RESOURCE AND MATERIAL EFFICIENCY

BEST PRACTICES GUIDELINE FOR CULTIVATION OF MICROALGAE ON NUTRIENT RICH DIGESTATE

PART 1:
BEST PRACTICES FOR THE TREATMENT AND PREPARATION OF NUTRIENT RICH DIGESTATE FOR ALGAL CULTIVATION

EDITION : MARCH 2020

Introduction
1

anaerobic digestion of waste

on the type of waste and its composition, as well as the quantity of waste treated and the operating temperature of the digester. Anaerobic digestion can be divided into six stages (Jayaseelan, 1997): hydrolysis of complex organic biopolymers into monomers; fermentation of amino acids and sugars; anaerobic oxidation of volatile fatty acids and alcohols; anaerobic oxidation of intermediary products such as volatile fatty acids; conversion of hydrogen to methane; conversion of acetate to methane. Commonly the AD process is divided into hydrolysis, acidogenesis, acetogenesis and methanogenesis stages. This process results in the production of biogas converted into electricity by a co-generator or directly used by injection after purification. The AD process also results in the production of a nutrient rich liquid digestate with a high dry matter content (Figure 1).

Digestate is often extremely rich in carbon, nitrogen, phosphorus and other macro and micronutrients (Papadimitriou et al., 2008; Tambone et al., 2017). All of these components are key factors in the growth and development of microalgae, which are aquatic photosynthetic microorganisms. When grown on wastewater or liquid digestate as a medium substrate, microalgae present a significant potential for the bioremediation (e.g. nitrogen and phosphorus uptake) of the mentioned waste (Fathi et al., 2013; Judd et al., 2015; Luo et al., 2017).

STEP 2 Fermentation
BACTERIA COMMUNITY
ANEROBIC DIGESTION

STEP 3 Anaerobic oxidation (volatile fatty acids and alcohols)

STEP 4 Anaerobic oxidation intermediary products such as volatile fatty acids

NUTRIENT RICH DIGESTATE

BIOGAZ PRODUCTION

BIOFERTILIZER FOR CROPS

ALG-AD PROJECT

NUTRIENT SOURCE FOR MICROALGAE CULTIVATION

PRODUCTION OF HIGH VALUE COMPOUNDS FOR ANIMAL FEED

Nutrient Content

Analysis of nitrogen, phosphorus and heavy metals also took place in this dilution/bestment trial. Nitrogen was relatively high in the control (i.e. 100% concentration of digestate) with a concentration of 8550 mg/L, which is expected for a nutrient rich digestate and this result was comparable with results from literature (Kumar et al., 2010; Marouf, 2015; Wang et al., 2010; Zhou et al., 2012). Values of total nitrogen in the 10% and 50% digestate mixture, which were significantly lower than the control, showed that diluting the digestate with water significantly affected the total nitrogen content of the digestate. On the other hand, the recalculated nitrogen content of the 10% digestate mixture was higher than the 10% digestate concentration (1925 mg/L and 809.6 mg/L respectively). This result showed that dilution allowed for some of the phosphate bound to the digestate to be available with regards to microalgal cultivation. Indeed, a modification of the process as simple as a shift of the N to P ratio, which is specific to the microalgal species, allowed for a significant increase in the growth of the microalgae (Gander and La Roche, 2002).

Figure 4: Phosphate (A) and nitrogen (B) content in 100% digestate control (in grey), 50% and 10% digestate (light blue) and recalculated values for 50% and 10% digestate (dark blue)

Parameter	Control	50%	10%
Phosphate (mg/L)	~1800	~1000	~800
Nitrogen (mg/L)	~8550	~1925	~809.6

Figure 5: Treatment and preparation of nutrient rich digestate for algal cultivation

Best Practice documents

Decision support tool

Scientific publications

Webinars recordings

All available on the **ALG-AD website**



Thank you



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