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**BIOSYSTEMS & FOOD
ENGINEERING
RESEARCH REVIEW No 28**



**UCD SCHOOL OF BIOSYSTEMS
AND FOOD ENGINEERING
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Editors: Tamíris da Costa and Junli Xu

Foreword

The Twenty Eighth Annual Research Review describes the ongoing research programme in the School of Biosystems and Food Engineering at University College Dublin over the academic year 2022/23, from the collective research body within the school comprising our academic staff, technical staff, research staff and our early-stage researchers.

A copy of this book is available to download from the UCD Research Repository at: <http://researchrepository.ucd.ie>.

The research programme covers two main focal areas: Food and Process Engineering as well as Energy and the Environment. Each of these areas is divided into sub-themes as indicated in the Table of Contents, which also includes the name of the research scholar (in bold); the title of the research and the nature of the research programme.

The review also highlights the award winners for presentational excellence at the 28th Annual Biosystems and Food Engineering Research Seminar, which was held online in virtual format on Thursday 16th March 2023. The awardees for 2023 are listed in the Appendix A.

The Editors gratefully acknowledge the dedicated work of the individual research scholars, their research supervisors and the financial support of research sponsors that enable this research review.

This review includes short papers prepared from the participants of the School's Taught Masters Programmes 22/23 as follows:

ME - Biosystems and Food Engineering <https://www.ucd.ie/engineer/engineering/biosystemsfood/index.html>

MEngSc – Food Engineering <https://www.ucd.ie/engineer/engineering/biosystemsfood/food.html>

MSc – Environmental Technology <https://www.ucd.ie/engineer/engineering/biosystemsfood/environmental.html>

MSc – Sustainable Energy and Green Technologies <https://www.ucd.ie/engineer/engineering/biosystemsfood/sustainable.html>

TABLE OF CONTENTS

Title Page	i
Foreword	ii
Table of Contents	iii

FOOD & PROCESS ENGINEERING

Food Engineering

Maniyar A , Monitoring the growth and lipid content of microalgae during cultivation in brewery wastewater (MSc Research)	1
Chaple S , Cold plasma interactions with food ingredients functionality (PhD)	2
Cheng Y , The influence of genetics and environment on the growth of <i>Listeria monocytogenes</i> in low temperatures (PhD)	3
Gandhi D , Effects of cold plasma treatment on barley flour with chickpea flour (MEngSc)	4
Dong G , Investigating the effects of different post-harvest treatments on allergens in Irish seaweed (PhD)	8
Shruti M , Influence of cold plasma on protein enriched wheat flour (MEngSc)	9
Viona R , Assessment of odour emissions from dairy wastewater (MEngSc)	13
Ren Y , Advanced application of Terahertz time-domain spectral technique for in-situ monitoring of microwave vacuum dehydration (PhD)	17
Hu Z , Isolation of proteins from potato (<i>Solanum tuberosum</i>) using ultrasound and enzyme-assisted extraction techniques (PhD)	18
Lucamante M , Microalgae for the bioremediation and valorisation of brewery effluents (MSc Research)	19
Serlini N , Understanding the ultrastructure of microalgae cell walls to enable biomass fractionation (PhD)	20
Shivakumar S , Microalgae cell-wall analysis and enzymatic disruption (PhD)	21
Barroug S , Chicken Juice a food-based model to assess the antimicrobial efficiency of three cold plasma approaches (PhD)	22
Fu Y , Investigating the dehydration feature of beef products with different pre-treatment during microwave vacuum dehydration using NIR-HSI and terahertz spectral analyses (MEngSc)	23
Noore S , Extraction yield and biological activity of phycobiliproteins from <i>Porphyridium purpureum</i> using atmospheric cold plasma discharge and jet systems (PhD)	27
Sun C , Growth of <i>Listeria monocytogenes</i> in mozzarella cheese (MEngSc)	28
Tang J , Influence of cavitation technologies, mainly hydrodynamic and ultrasonic cavitation, on structure and functional properties of pea protein (PhD)	32
Yang K , Identification of mushroom browning patterns using a portable hyperspectral imaging camera combined with machine learning techniques (PhD)	33
Zhang Z , Risk assessment of <i>Listeria monocytogenes</i> in butter (MEngSc)	34
Rajappa P , Co-cultivating algae and fungi for food production (MEngSc)	38
Ravichandran P , Co-cultivation of microalgae and fungi (MEngSc)	39

Anilkumar A , Predicting the soluble solid content and moisture content in apples by vacuum drying techniques using terahertz time domain spectroscopy (MEngSc)	40
Fiore V , Controlling spores in dairy powders: Spore population during processing stages of skim milk powder (PhD)	44
Li Q , Predicting wheat gluten concentrations in potato starch using GPR and SVM models built by terahertz time-domain spectroscopy (PhD)	48
Lyu H , Access the effects of fermentation time and point of grass silage bale on grass quality (ME)	49
Dong W , Haddock protein recovery by ultrasound-assist isoelectric solubilisation and alcalase hydrolysis (PhD)	53
Zhu X , Hydrodynamic cavitation for brown seaweed in a cascading biorefinery model for laminarin, alginate and protein extraction (PhD)	57

Imaging, Risk Assessment, Traceability

Canga E , Investigating the microplastics aging under some conditions and the fate of microplastics in food biological systems (PhD)	58
Ferreira R , The effect of UV ageing on spectral properties of polystyrene microplastics (PhD)	62
Fhuaráin A , An investigation into the influence of sample presentation in the measurement of milk powder using attenuated total reflectance (ATR) mid-infrared (MIR) spectroscopy and principal component analysis (PCA) (PhD)	66
Govil K , Investigating the release of microplastics from bakeware during oven heating (MEngSc)	67
Nic S , Optimising plasma functionalised liquids for the treatment and control of orthopaedic implant infections (PhD)	71
Özdoğan G , Feasibility of spectral imaging and data fusion for classification of Turkish wheat (PhD)	72
Najafpour T , Estimation of chlorophyll content in coffee arabica leaves based on hyperspectral imaging analysis (PhD)	73
Xie J , Development of a microplastic detection framework using optical photothermal infrared spectroscopy (PhD)	76
Yang C , Spectral imaging for identification of polymer degradation (PhD)	80
Zhao Q , Automatic recognition and grading of strawberries using portable hyperspectral imaging camera: A preliminary study (MSc)	81

Biosystems

Kehoe C , Protocols for the digital collection of data for decision making in crop production (PhD)	85
Ma Q , Microalgae bioremediation of brewery wastewater (PhD)	89
Wang S , The effects of ultrasound treatment with different frequency on physicochemical and functional properties of fava bean proteins (PhD)	90
Ramireddy L , Agriplasma: Non-thermal air plasma treatment of multispecies swards seeds for reduction of greenhouse gas emissions (PhD)	91
Macharaja R , Economic and feasibility analysis of renewable energy installation in medium-scale distilleries (ME)	92

Macartan B , Effects of feed additives on ammonia and greenhouse gas emissions during manure storage (PhD)	96
Nuwayhid S , Cultivation of microalgae on brewery waste (ME)	100
Bhatia V , Ranking strategy for microbial hazards associated with RTE fresh produce following the use of recycled wastewater for irrigation (PhD)	101
Putsakum G , Low-intensity pulsed electric field as a pre-treatment for fresh blackberries (PhD)	102

ENERGY & THE ENVIRONMENT

Environmental Technology, Modelling, Risk Assessment

Bai F , Comparison of portable instrumentation for spectral imaging of bacteria (MSc Research)	103
Roosmalen R , Development of a whole cell exclusion biosensor which is able to detect a whole range of analytes present in grass leachate (PhD)	107
Charisis C , Deep learning techniques for yield prediction in multi-domain mushroom production environments (PhD)	108
O’Riordan L , Developing a policy landscape to support the development of a grass to protein biorefinery centre in Ireland (MSc)	109
Pollard P , Ensiled seaweed as an alternative biorefinery input substrate (PhD)	113
Visentin A , Development of contaminant removal strains for the downstream purification processes in a green biorefinery (PhD)	114
Leelasuksun W , Biobeo: Food loop, innovative education for the bioeconomy (MSc)	115
Anand S , Biobeo (interconnectedness), innovative education for the bio-economy (MSc)	119
Das A , Forestry and outdoor learning in Europe (MEngSc)	123
Yuan Z , Human health risk assessment of microplastics through seafood products (PhD)	127
Vazquez V , Human exposure to BPA analogues through food (MSc Research)	128
Cerca M , Strategies for biomass supply systems in the context of an emerging circular bioeconomy: Inclusion of human dimensions in sustainability transformations (PhD)	132
Vance C , Understanding potential environmental and social impacts of Irish seaweed production through life cycle assessment (PhD)	133
Wang X , Comparative risk assessment study on bisphenol A (BPA) through meat products (PhD)	134

LCA, Sustainable Agriculture & Soil Resources

Vergara L , The environmental impact of climate mitigation strategies over a commercial Irish dairy farm (PhD)	135
Shi L , Predicting soil carbon sequestration potential on Irish soils from spectral data (PhD)	136
Kabiri S , Soil organic carbon mapping with semi-supervised learning on VIS-NIR hyperspectral images (PhD)	137
Jamil U , Exploration of geopolrisk to understand antimony resource depletion for the eu thermoelectrics market (PhD)	138

Sustainable Energy and Green Technology

Li Y , Valorisation of dairy waste streams using microalgae (PhD)	142
Brooke F , Microalgae application of brewery solid waste (PhD)	143
Iyohaiyoke O , Economic solutions for living off the grid through the integration of various renewable energy sources (MSc)	144
Finneran M , Literature and market review of mycelium-based biocomposite material (MSc)	148
Rahimi M , Lipid extraction from wet microalgae biomass for the production of omega-3 rich animal feed (PhD)	152
Xiao M , Microalgae bioremediation of dairy wastewater (PhD)	153
Bey S , Enhancing food safety of fresh poultry meat using cold plasma technology and natural compounds (MSc Research)	154

Appendices

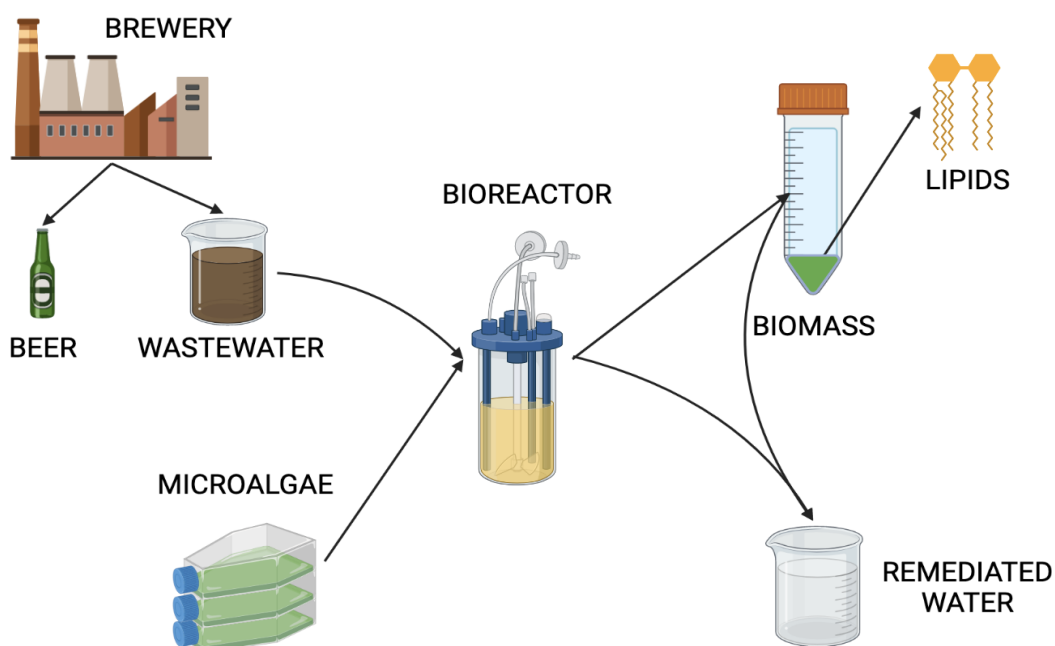
A. Award winners	159
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Project Title: Monitoring the growth and lipid content of microalgae during cultivation in brewery wastewater

Project Leader: Dr. Ronald Halim

Abstract

Brewery wastewater can pose major environmental concerns if released untreated and conventional chemical methods also present environmental and cost issues. Microalgae was cultivated to study its potential as a novel method of remediating and valorising brewery wastewater. This can be accomplished by production of microalgal biomass, which can then be harvested for lipid extraction. It was found, based on preliminary results, that microalgae were able to grow on wastewater. This was concluded based on measurements of optical density at 750 nm of samples collected over three weeks as well as the final measurement of biomass generated after centrifugation and freeze-drying. The bioremediation potential will be assessed by measuring nitrate, phosphate, and ammonium removal, as well as lipid yield (Bligh and Dyer extraction) and dry weight measurements of the biomass generated.



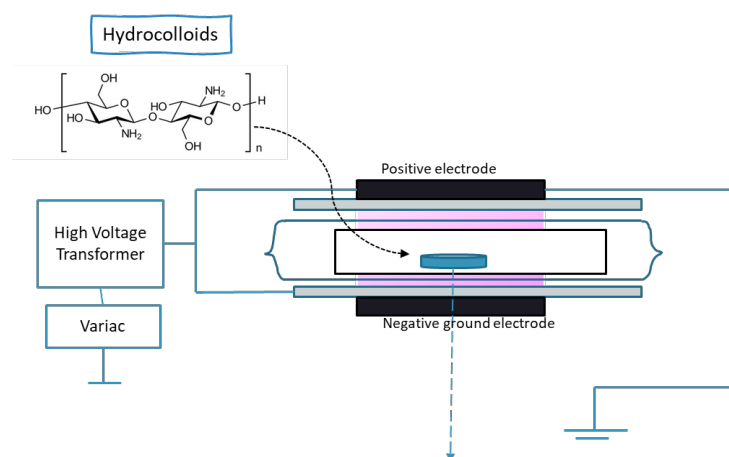
Sonal Chaple, B.Tech., M.Tech.

Project Title: Cold Plasma interactions with food ingredients functionality

Project Leader: Prof. Paula Bourke

Abstract

The use of chemical treatment for modification or improvement of product functionality is a very common practice. These modifications often lead to label amendments. The use of chemical can also result in chemical waste giving environmental concerns. Also, consumer awareness about food safety has increased tremendously in recent years. Hence, the food industry is continuously looking for innovative alternatives to current processing technologies tuned to add functionality to diverse food systems. Atmospheric cold plasma (ACP) has emerged as a novel processing technology, with demonstrated efficiencies in microbial inactivation. ACP can be designed to be cost-effective, sustainable, non-thermal and offer system and process design versatility. However, studies on the effects of ACP and potential to modify the functional properties of foods are sparse. The objective of this study is to determine the effect of ACP on physico-chemical and functional properties of wheat flour. In this study, hydrocolloids (Chitosan, xanthan gum, gum arabic, xanthan gum and sodium alginate) were subjected to a dielectric barrier discharge (DBD) contained plasma reactor for a range of treatment times (5–30 min) at 80 kV. An increase in L*(lightness) and b* value was observed for gum arabic samples with plasma treatment. The increase in b* value can be due to an increase in porosity due to etching of plasma-treated gum. The bulk compressibility of chitosan powder was found to decrease with application of cold plasma. The decrease in bulk compressibility can be attributed to the etching effect of the cold plasma on the surface. Shear properties will provide important information such as whether the powder will flow through the process. With the plasma treatment the flowability characteristics of the powder have changed, making it free-flowing. High cohesive strength and unconfined yield of powder result in lower Ffc giving it the poor ability to pass through hopper. A broad peak (observed at $2\theta=18.97$) without any intense peak suggests an amorphous nature. A change in peak intensity was observed with increasing treatment time. Depolymerization or change in particle size distribution can affect peak intensity.



Selected Recent Publications

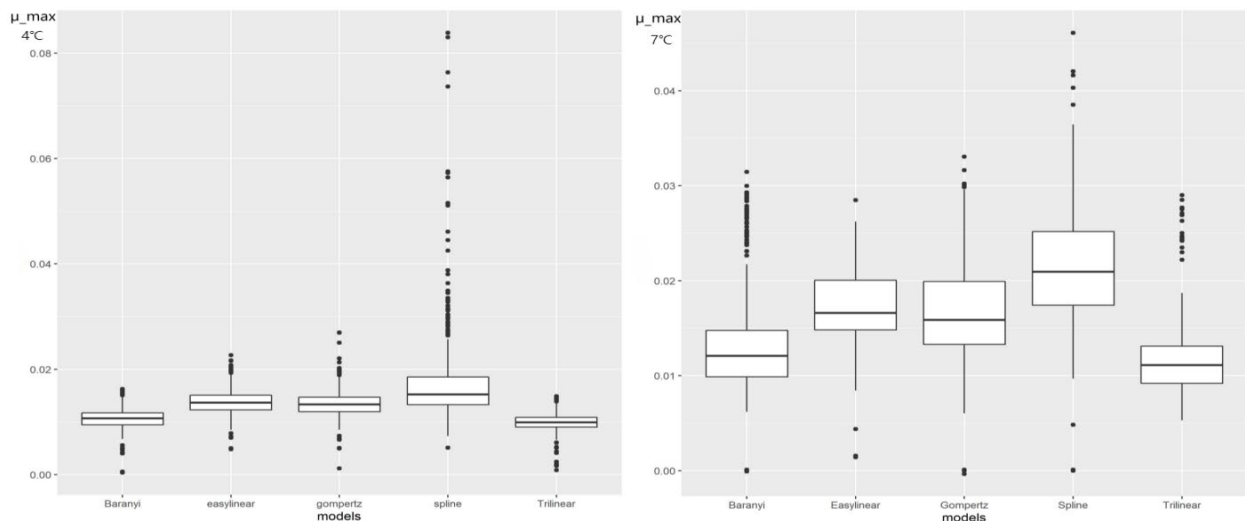
Barroug, S., Chaple, S. and Bourke, P., (2021). Combination of Natural Compounds With Novel Non-thermal Technologies for Poultry Products: A. *Natural Compounds in Food Safety and Preservation*.

Project Title: The influence of genetics and environment on the growth of *Listeria monocytogenes* in low temperatures

Project Leader: Prof. Francis Butler

Abstract

Listeria monocytogenes is one of the most virulent foodborne pathogens. *L. monocytogenes* can grow at temperatures as low as -0.4°C , which allows the organism to grow to infectious doses even at low temperatures. Nowadays, the value of whole genome information in the study of the stress responses of *L. monocytogenes* has already been reported. Therefore, the purpose of this study is to choose a suitable model to fit the growth of *L. monocytogenes* at low temperatures and obtain the growth rate, and to explore the association between the genetic factors, environment, and the growth of *L. monocytogenes* in low temperatures. 150 *L. monocytogenes* strains and their whole genome sequence data were used in this study. The growth data of 150 strains were measured at 4°C and 7°C respectively, and five models were used to fit their growth curves. The criteria for suitable fitted models are currently under consideration. The highest growth rates generated by the five fitted models are shown in the figure below. According to the whole genome sequence data of the sample strains, multi-site sequence typing was used to classify the strains, and gene alignment was used to determine the content of low temperature resistance genes in the strains. At present, the gene alignment of 11 resistance genes has been carried out, and it is found that all strains carry these 11 resistance genes. Therefore, the number of resistance genes carried is not a factor affecting the growth of *L. monocytogenes* in a low-temperature environment. In future work, regression analysis will be used to determine the association of environmental factors and sequence types with the growth rate of *L. monocytogenes* in low temperature environments.



EFFECTS OF COLD PLASMA TREATMENT ON BARLEY FLOUR WITH CHICKPEA FLOUR

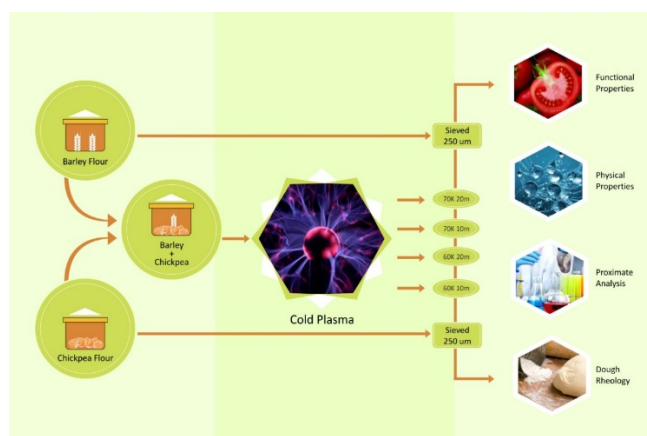
Divya Gandhi, Paula Bourke

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

Recently, atmospheric cold plasma (ACP) based on dielectric barrier discharge (DBD) has emerged as an emerging food development technology as well as a novel and promising plasma technology (a non-destructive preservation technology) that has potential applications in the food industry. In this context, the current study provides an overview of recent advances in ACP application and its impact on the functional, physicochemical, and sensory attributes of barley flour products. The treatment involves subjecting food products to a non-thermal, low-pressure plasma generated by electrical discharge, causing various physical and chemical changes in the food. Particular interest in cold plasma treatment research is the potential impact of this technology on the functional and nutritional properties of barley flour, chickpea flour, and mixtures of the two. In this study, we aim to further improve the nutritional value of barley flour by incorporating a protein-rich ingredient, chickpea flour. We will then subject this flour mixture to cold plasma treatment to assess its impact on functional and nutritional properties.

Graphical Abstract



Introduction

The analysis of non-traditional food processing methods is usually conducted from a microbiological viewpoint, however though its impact on food chemistry. Flour enhancers, dough conditioners, or bleaching agents, such as oxidising agents and enzymes, are commonly added. However, chemical oxidising agents have drawbacks, such as the potential introduction of toxic effects to the food chain. Enzymes are expensive and can cause structural modifications. In contrast, cold plasma has been demonstrated to influence the chemical constituents of food, impacting its quality. Barley flour is an ingredient widely used in diverse food applications, including baked goods, snacks, and beverages. It is abundant in dietary fibre, vitamins, and minerals and exhibits functional properties such as water retention, gelation, and emulsification. The potential of cold plasma treatment for enhancing the physicochemical and functional properties of cereal flour has been investigated in several studies (Jannasch *et al* 2020). These investigations have revealed promising outcomes, highlighting the efficacy of cold plasma treatment as a tool for improving the quality

and functionality of barley flour in various food products (Kermasha *et al* 1993). In the past, different techniques, such as microwave, autoclave steaming, roasting, toasting, and infrared heating, have been evaluated to modify and stabilise the functional properties of barley flour.

Cold plasma technology can modify the chemical makeup of wheat flour by inducing radical and ozone propagation oxidation, which in turn alters its functional properties. Nonetheless, it is essential to establish the extent of these changes and its control for the enhancement of the flour's functionality. To this end, the present investigation aims to evaluate the impact of low doses of cold plasma treatment on the microbial load, chemical composition (protein and lipid), and dough rheology of wheat flour. A high voltage treatment level was chosen to minimise secondary effects such as the production of oxidation products that could influence the aroma.

The objective of this study was to examine how cold plasma treatment affects barley flour, in comparison to chickpea added high protein barley flour, find the ideal combination of cereal-legume flour composition by using chickpea and barley mixture and analyse how cold plasma treatment affects the physical properties, functional properties, dough rheology of this flour mixture.

Materials and Methods

Sample Collection

Samples of Barley and chickpea flours were procured from Evergreen health food store.

Blending of Samples

The barley flour used in the study was standardised to a particle size of 250 microns by sieving. This sieving process was carried out using a sieving machine (Endecotts Ltd., London, England) in the laboratory of the Food Science building, UCD.

Atmospheric Cold Plasma Treatment Parameters

For the treatment of the barley flour, 15 g of flour samples were placed in petri dishes and treated with four different parameters using an atmospheric cold plasma (ACP) prototype referred from previous work (Chaple and Sarangapani, 2020). The parameters included a 20-minute treatment at 70 kV (sample BF7020), a 10-minute treatment at 70 kV (sample BF7010), a 20-minute treatment at 60 kV (sample BF6020), a 10-minute treatment at 60 kV (sample BF6010).

Atmospheric Cold Plasma Set-up

The ACP source was powered by a step-up transformer (Phoenix Technologies, Inc., MD, USA, is the transformer supplier), which regulated the input voltage using a variation. The DBD unit included two circular aluminium electrodes (with an outer diameter of 158 mm) and two polypropylene dielectric layers (the gap between the electrodes is >3 cm). The input voltage to the transformer was regulated to produce an operating frequency and applied powers of 200 W/cm² and 212 W/cm², respectively. The barley flour samples were placed in polyethylene terephthalate and sealed with a high-barrier polythene film. The treatment was carried out under an atmospheric air condition of 40 ± 10% relative humidity and temperature 20 ± 2°C, which was measured using a humidity-temperature probe connected to a data logger (Testo 176T2, Testo Ltd., UK). Treatments were done in triplicates. The ozone concentration inside the treatment chamber was ranging from 0.03 to 0.05 ppm, measured during treatment using an ozone metre. The treated samples were stored at room temperature for 24 hours post treatment. The study examined the physical, proximate, functional properties of barley flour and chickpea flour separately when BF7020, BF7010, BF6020, BF6010 exposed to plasma and Controlled (untreated sample).

Physical analysis

Colour intensity of the composite flour blends will be measured using a Konica Minolta Colour Measuring System (Chroma meter CR-410, Minolta LTD Japan) as described by Ahmed and Hussein (2014).

Bulk Density (BD) of the sample will be determined using the method described by Onwuka (2005).

Functional

Properties

Functional Properties which will be analysed (Mohammed *et al* 2014) are:

Swelling capacity: The supernatant will be decanted and sedimented paste will be weighed.

Swelling capacity (g/g) = (Weight of sediment (g))/ (Dry weight of the sample (g))

Solubility: The solubility of the sample will be calculated by equation (the value obtained multiplied with 2 because only 5 ml of the supernatant will be taken out from the initial volume of 10 ml water).

solubility (g/g) = (Weight of soluble sample (g))/(Weight of sample (g))×2

Water absorption capacity: The method described by Onwuka (2005) will be used.

Oil absorption capacity: The method described by Onwuka (2005) will be used. Dispersibility will be analysed by the method described by Kulkarni, Kulkarni, and Ingle (1991).

Emulsion activity (EC) will be determined using the method of Kaushal *et al* (2012).

Emulsion stability will be determined using the method of Kaushal *et al* (2012).

Foam capacity and foam stability will be determined according to the method described by Onwuka (2015).

Least gelation concentration (LGC) of flours will be determined by the method of Sathe, Desphande, and Salunkhe (1982).

Proximate analysis tests

Carbohydrate, sugar, fat, protein, fibre, ash will be carried out by the AOAC methods. Out of them, protein and fat analysis will be analysed in the Science lab.

Dough Properties

It will be analysed are MicroVisco-Amylo-graph and Farinograph (Brabender, Duisburg, Germany) & Rheometer.

Results and Discussion

Expected Results

The treatment was effective in creating changes in the lipid components of the flour. Cold plasma treatment was found to result in a voltage and treatment time dependent reduction in non-starch FFAs and phospholipids and progression of oxidation was evidenced by a significant increase in PV and n-hexanal concentration at all levels of treatment, suggesting that cold plasma is resulting in an acceleration of flour oxidation. The cold plasma treatment did have a significant effect on the free fatty acid and phospholipid complement of the cereal flour (Bahrami *et al* 2016).

Conclusions

Not concluded.

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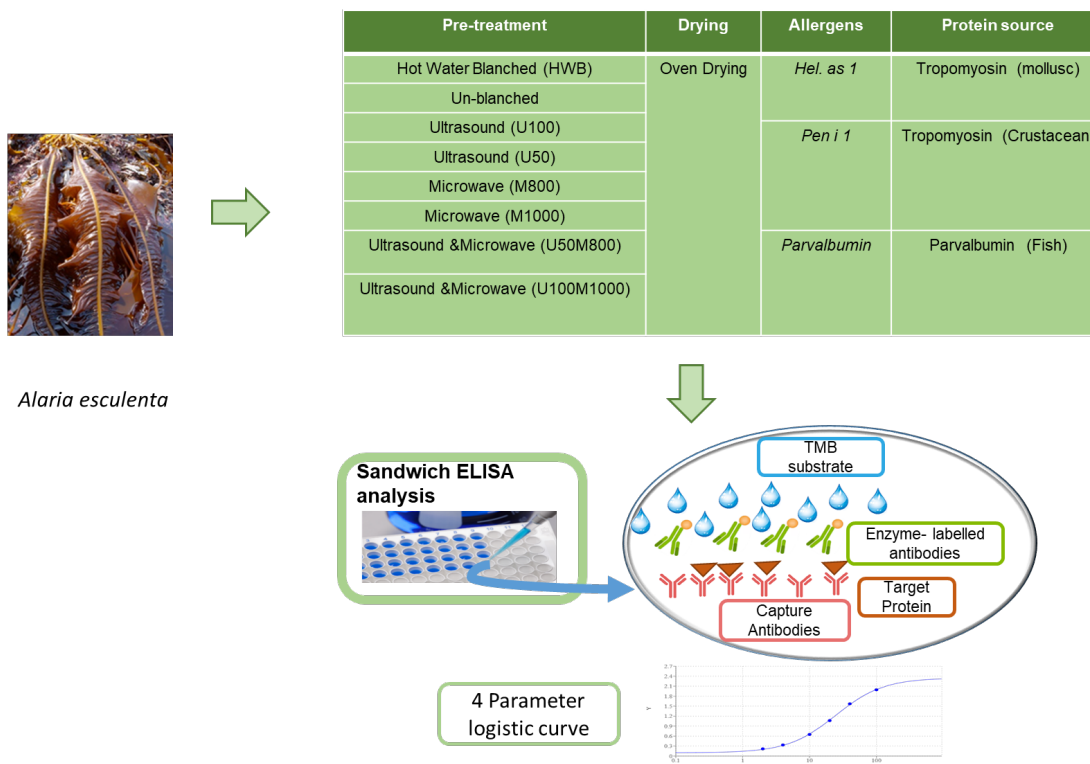
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Project Title: Investigating the effects of different post-harvest treatments on allergens in Irish seaweed

Project Leader: Da-Wen Sun, Brijesh K. Tiwari

Abstract

Seaweed is a promising alternative protein source that has not been fully exploited. According to the European food safety authority, allergenicity of the novel food should be labelled for food safety concern. This study aimed to investigate the different post-harvest treatments on allergens in Irish seaweed. Seven pre-treatments were carried out on *Alaria esculenta* including conventional hot water blanching (HWB), a novel ultrasonic blanching and microwave blanching, alone or in combination. Three allergens from two main types of allergenic protein were investigated using sandwich ELISA analysis, namely, tropomyosin (Hel as 1 from mollusc and Pen i 1 from crustacean) and parvalbumin (from fish). A four-parameter logistic model was used to calculate the allergen concentrations based on the standard curve. One-way ANOVA illustrates the differences between different processing methods. In general, pretreatment significantly reduced the allergenicity of Parvalbumin and Hel. as 1 in *Alaria esculenta* ($p < 0.05$). The most effective pretreatment was a combination of ultrasound and microwave at 100% amplitude and 1000 W, which resulted in a 94.96% reduction of Parvalbumin content in *Alaria esculenta*. Overall, these findings suggest that ultrasound treatments, alone or in combination with microwave, can effectively reduce the allergenicity of *Alaria esculenta*.



INFLUENCE OF COLD PLASMA ON PROTEIN ENRICHED WHEAT FLOUR

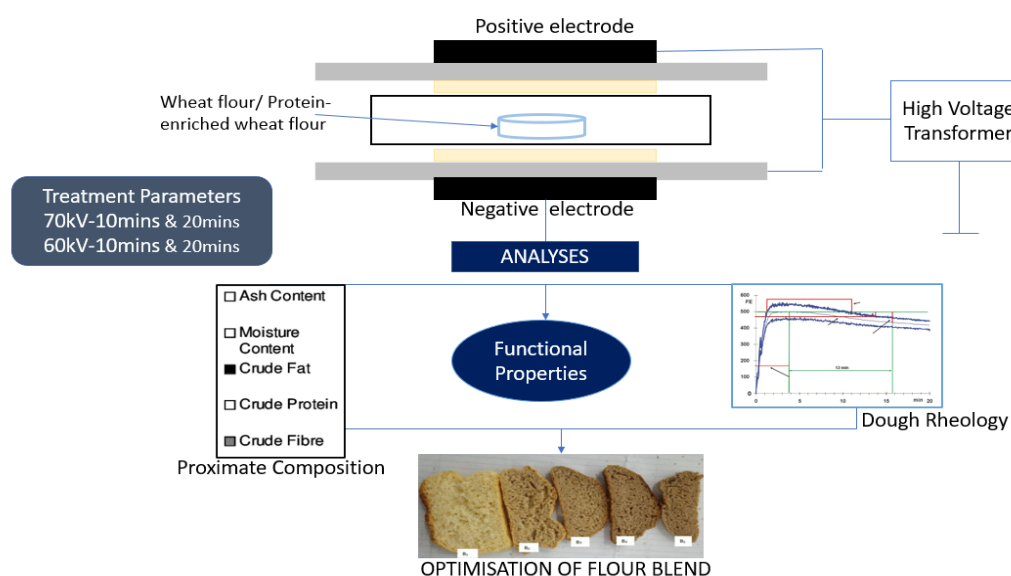
Menon Shruti, Bourke Paula

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

Atmospheric pressure cold plasma (ACP) has the potential to alter physical surface properties and biological chemistry. ACP or atmospheric cold plasma has become a novel processing technique with proven efficacies in microbial inactivation. The objective of this study is to determine the effect of ACP on physio-chemical and functional properties of protein enriched wheat flour and compare it with the effects on wheat flour alone. In this work, wheat flour and chickpea flour were individually and in combination (10%, 20% and 30% mixing ratio) were treated to a dielectric barrier discharge (DBD) atmospheric pressure cold plasma reactor at a high voltage of 60 and 70kV, for 10 and 20 mins of exposure time. Following analysis of the preliminary results obtained, a flour mixing ratio was optimized. According to an assumption based on earlier research articles, applying a plasma treatment may result in improved hydration and dough rheology. The findings of this study provide valuable insights on the possible advantages and uses of cold plasma treatment in terms of quality and safety of flour blends.

Graphical Abstract



The overall aim of this study is to analyse the influence of cold plasma on the flour blends and optimise according to the results obtained.

Materials and Methods

Raw material acquisition

Organic wheat flour was sourced from Ballybrado Ltd. Ireland and organic chickpea flour was purchased from Bulkfoods, Ireland. All chemicals and reagents used were of analytical grade.

Preparation of samples

The wheat and chickpea flours were passed through British Sieve Standards BS No.250 mesh sieve to maintain a standard particle size. These particles were then subjected to DBD-ACP treatment with four treatment parameters- 60kV for 10 and 20 minutes, 70kV for 10 and 20 minutes.

Blending of flour

Each flour was passed through British Sieve Standards BS No.250 mesh sieve and composite blends of wheat flour and chickpea flour were mixed according to three ratios 10%, 20% and 30%. Plasma treated wheat flour for 10 and 20 minutes is termed as PTWF-10/60, PTWF-20/60, PTWF-10/70, PTWF-20/70 and plasma treated mixture of chickpea flour and wheat flour for 10 and 20 minutes is termed as PTMF-10/60, PTMF-20/60 and PTMF-10/70 and PTMF-10/70.

Treatment of samples

15g of each sample was weighed separately in a petri dish and placed in polypropylene boxes which were then sealed within a packaging film (Cryovac, Ireland) (Sarangapani *et al* 2017). All samples were subjected to plasma treatment using an output discharge voltage of 60 and 70kV, with treatment duration of 10 and 20 minutes. An atmospheric air plasma reactor, based on a dielectric barrier discharge (DBD) design was used in this study. Briefly, plasma is generated between two electrodes using two acrylic dielectric layers. The applied voltage to the electrode was provided by a high voltage transformer (Phenix Technologies, Inc., MD, USA). The treated samples were stored at room temperature for 24 h post treatment storage time.

Sample analyses

Proximate Composition

The proximate composition of the flour samples and blends are done according to the AOAC (2010) method.

Determination of Protein Content

The Kjeldahl method is used to determine the organic nitrogen content in the flour sample. 1g sample is weighed and transferred to Kjeldahl digestion tube. The tubes were heated, and the digest cooled, the distilled NH₃ was trapped until 150ml of distillate formed and the colour change from red to green observed.

Determination of crude fibre content

The crude fibre was estimated by subjecting the flour sample to acid and alkali digestion (AOAC, 2010). Defatted sample was weighed and boiled in 200 ml of 1.25 % sulphuric acid for 30 min. The sample was passed through a muslin cloth and the residue washed with distilled water until no acid residues remain. The retentate was collected and boiled in 200 ml of 1.25 % sodium hydroxide solution for 30 min. The sample was again passed through muslin cloth followed by repeated washing. Remove the residue and transfer to a pre-weighed crucible W₁. Dry the residue for 2 h at 130 ± 2°C followed by cooling it in a desiccator and weigh as W₂. Ignite the sample for 30 min at 600 °C in a muffle furnace. Cool the contents in a desiccator and weigh W₃. The obtained value was substituted in formula.

$$\% \text{ of crude fibre} = \frac{(W_2 - W_1) - (W_3 - W_1) \times 100}{\text{Weight of sample}}$$

Determination of Fat Content

Lipid fraction from the food is extracted using a non-polar solvent and the solvent is subsequently evaporated and the amount of fat extracted is determined gravimetrically AOAC International (1995). The Soxhlet method was used to determine the fat content and the formula used is,
 $\% \text{Fat} = (W_3 - W_1) \times 100 / W_2$

Determination of ash content

Was performed as described by P Williams *et al.*,1981. Reference ash content analysis was performed in triplicate in the Perten Inframatic 86 series NIR with ash kit. Samples were analysed on the instrument and slope and bias of the ash results were calculated. Corrections were verified by reanalysing a separate sample set.

Functional Properties

Water absorption capacity

About 1 g of flour sample was weighed into a 15 ml centrifuge tube and suspended in 10 ml of water. The sample was allowed to stand for 30 min at ambient temperature 30 ± 2 °C and centrifuged at 3000rpm for 30 min. The volume of free water is transferred to a measuring cylinder and is then recorded according to the method stated in Onwuka (2005).

Oil absorption capacity

As per Onwuka (2005), 1.0 g of flour sample was weighed into a 15 ml centrifuge tube and suspended in 10 ml of sunflower oil. The sample was allowed to stand for 30 min at ambient temperature 30 ± 2 °C and centrifuged at 300 rpm for 30 min. The volume of free oil is transferred to a measuring cylinder and is then recorded.

Foaming capacity and foaming stability

1.0 g flour sample was added to 50 mL distilled water at 30 ± 2 °C in a graduated cylinder.

$$\text{Foam capacity (\%)} = \frac{((\text{Volume of foam AW} - \text{Volume of foam BW}) \times 100)}{\text{Volume of Foam BW}}$$

Where, AW = after whipping, BW = before whipping. The suspension was mixed and shaken for 5 min to foam. The volume of foam after 30s whipping was expressed as foam capacity using the formula, foam volume according to the method in Onwuka (2005).

Emulsion activity and Emulsion stability

1 g of raw or processed flours was mixed with 10 ml of distilled water and 10 ml of vegetable oil in a calibrated centrifuge tube and shaken for 15 min. The emulsion was centrifuged at 2000 g for 30 min, and the ratio of the height of the emulsion layer to the total height of the mixture was calculated as emulsion activity in percentage (Kaushal *et al* 2012). The emulsion stability was estimated after heating the emulsion contained in a calibrated centrifuged tube at 80 °C for 30 min in a water-bath, cooling for 15 min under running tap water and centrifuging at $2000 \times g$ for 15 min. The emulsion stability expressed as percentage was calculated as the ratio of the height of emulsified layer to the total height of the mixture.

Swelling capacity

The swelling capacity of the flour sample was determined by dissolving 0.5 g of sample in 10 ml of distilled water and heating for 30 min at 60 °C with continuous gentle mixing to prevent the disintegration of starch. The acquired sample solution was cooled to 25 °C and centrifuged at 1600 rpm for 15 min. The supernatant was decanted and sedimented paste was weighed.

$$\text{Swelling capacity } \left(\frac{g}{g} \right) = \frac{\text{Weight of Sediment (g)}}{\text{Dry weight of the sample (g)}}$$

Dispersibility

The dispersibility of the flour sample was estimated by employing Ramashia *et al.*, 2017 method. 10 g of the flour was measured in 100 ml of a graduated measuring cylinder. The distilled water was added up to the mark and kept aside for 3 h without mixing. The volume of the precipitated particles was recorded and substituted in formula,

$$\text{Dispersibility (\%)} = 100 - \text{volume of sedimented particles}$$

Least gelation concentration

The least gelation concentration of the flour sample was estimated by employing Ehimen R. Ohizua *et al* (2017) method. The flour dispersions of 4%, 6%, 8%, 12% (w/v) prepared in 5 mL distilled water was heated at 90 °C for 1 h in water bath. The contents were cooled under tap water and kept for 2 h at 10 ± 2 °C. The least gelation concentration was determined as that concentration when the sample from inverted tube did not slip.

Result

The preliminary results are incomplete as the experiment is still under study and the statistical data analyses is not compiled. Based on the results obtained to date, the water absorption capacity of the wheat flour decreased while oil absorption capacity of wheat flour increased with increasing voltage and exposure time. The swelling capacity of the treated wheat flour decreased and there was no significant change in the emulsion activity of the wheat flour among the treated samples.

The gluten content in the final optimised mixture is expected to be less while the overall protein content is expected to increase, and the dough may be stronger resulting in enhanced bakery applications.

Conclusion

The plasma reactive species improve the hydration and functional properties of both wheat and chickpea flour. The plasma reactive species known to oxidize the protein chains and alter starch structure mainly amylose and amylopectin, resulting in changes in the flour functionality and dough rheology. As the major issue with protein enrichment by plant-based alternatives like chickpea flour is their low functionality. However, plasma as a pre-treatment may enhance their functionality and their compatibility for mixing in wheat flour. Thus, treated flour may result in a higher hydration properties and improved dough structures, providing scope for the nutrient rich products. The enrichment of protein from the chickpea flour results in an optimised ratio of flour blend which can be used for development of products.

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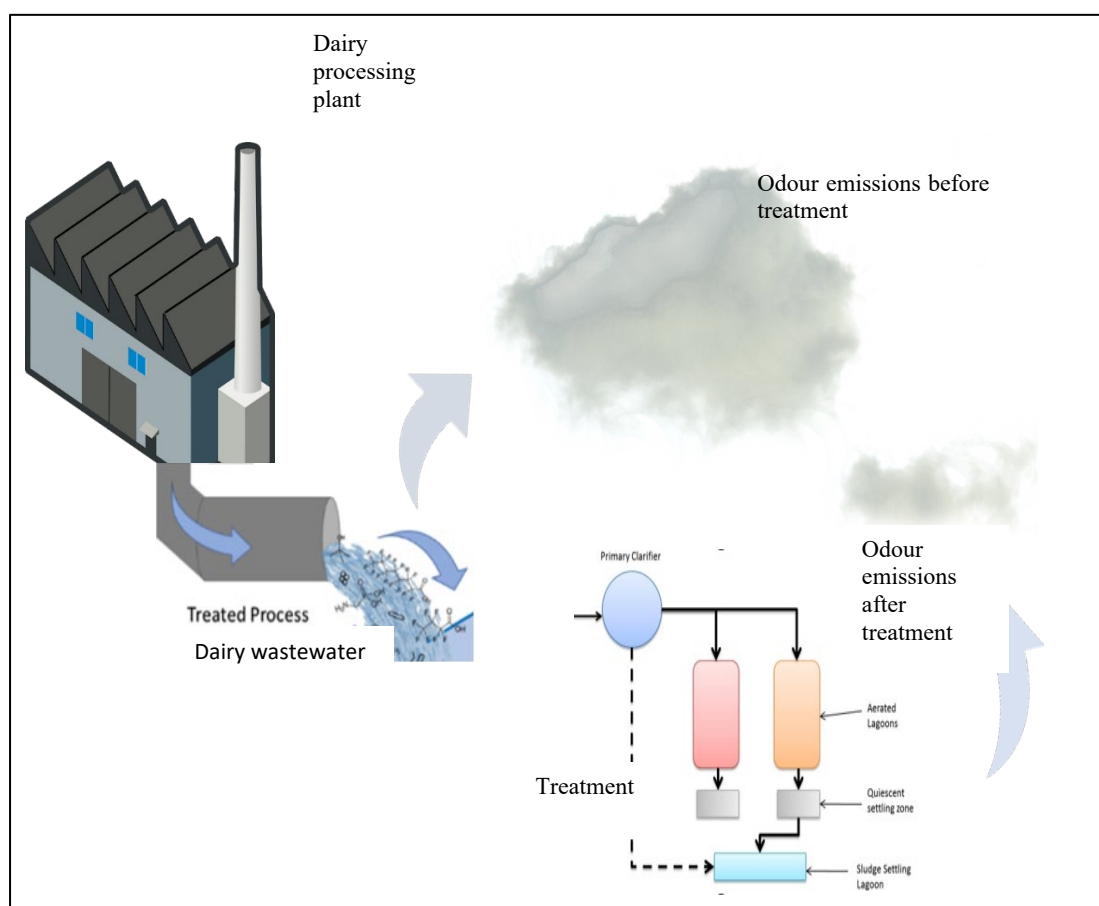
ASSESSMENT OF ODOUR EMISSIONS FROM DAIRY WASTEWATER

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Abstract

Dairy wastewater is a major source of odour emissions due to the presence of organic matter and nutrients providing a suitable environment for the growth of microorganisms that produce volatile organic compounds. This study aims to evaluate the odour emissions from dairy wastewater using both sensorial techniques and air dispersion modelling and assess the effectiveness of treatment technologies in odour reduction. The methodology involves standard dynamic sampling to collect samples, conducting sensory analysis using olfactometer tests to measure concentration, as well as using AERMOD air dispersion modelling to predict the dispersion of odours in the surrounding areas. The results of this study will provide insight into the potential impact of dairy wastewater odour emissions on the environment, as well as help identify effective treatment technologies for odour reduction. The study is limited to the dairy industry in Ireland, and the expected results may vary depending on the specific treatment technologies used. Overall, this study provides valuable information on assessing and managing odour emissions from dairy wastewater, which is essential for sustainable and responsible management of the dairy industry.



Introduction

Due to the high demand for dairy products, the projected increase in milk output is 1.6% by 2029. Therefore, this industry is one of the largest producers of industrial wastewater in the food industry (Kolev 2017). According to European Directives 91/2717/EEC and 97/771/ECC (CEN 2003), wastewater from dairy factories in Europe must receive special treatment, and its direct discharge into surface water is not permitted. Dairy effluent has been treated using a variety of methods, such as membrane technology, coagulation-flocculation, biological aerobic and anaerobic processes, and others (Ahmad *et al* 2019). These methods can lower the organic load such as chemical oxygen demand (COD), and other pollutants in wastewater, such as total suspended solids (TSS) and ammonia. Unfortunately, these treatments do not eliminate odours (Elia *et al* 2023).

Assessment of odours from the industry is greatly needed to limit pollution caused. Odours represent a major fraction of public complaints related to air pollution as they affect the quality of life and can cause physical symptoms and clear adverse effects on human health (Ranzato *et al* 2012). Odorants in the air can be hard to assess and regulate because in ambient air, they can be a large number and often in hard-to-detected concentrations. In addition, their intensity changes depending on the time of day and meteorological conditions. Odour perception is also subjective. To identify these odours, the odour concentration released at emission sources should be measured and evaluated at the receptors to establish a link between emissions and impacts (Ranzato *et al* 2012).

Several studies have been conducted in Ireland to investigate the sources and impacts of odour emissions from dairy wastewater. (Ahmad *et al* 2019) found that the main sources of odour emissions in dairy wastewater treatment plants were anaerobic digestion and aeration tanks. The study also showed that several factors, including the type and concentration of organic compounds, temperature, pH, and the presence of nutrients such as nitrogen and phosphorus, influenced emissions. (Conti *et al* 2020) evaluated the impact of odour emissions on nearby communities and highlighted the need for effective management strategies to mitigate odour emissions and protect public health. To address this issue, The Irish Environmental Protection Agency has developed guidelines for the management and treatment of dairy wastewater (Ireland 2001), however, they do not give a solution for elimination.

Odours are commonly measured with sensorial techniques, based on the detection of odours of the human nose. Sensorial techniques determine the characteristics of odorants in terms of the sensations that they cause and the threshold of perception. The dynamic dilution olfactometry technique is based on the employment of a trained human panellist and a dilution instrument: the olfactometer. It is used to mix gaseous samples with odourless air at several, predetermined ratios. The assessor smells the mixed air and gives a response in terms of perception or not. The technique allows the measurement of sample odour in odour units per cubic metre of gaseous sample and should follow the standard procedure described by EN13725 (CEN 2003). This is a standard method for measuring odour emissions. AERMOD is a steady-state dispersion model developed by the American Meteorological Society and U.S. EPA due to the consistency of meteorological conditions during the modelling period. This model is designed for stationary sources and emissions. In addition, AERMOD also can handle multiple point, area, and volume sources.

The objective of this study is to assess odour emissions from dairy wastewater using sensorial techniques and air dispersion modelling AERMOD, and the efficiency of treatment technologies in odour reduction.

Materials and methods

In this study, we analyse the efficiency of waste treatment systems of seven dairy processing plants in Ireland. All process wastewater is collected in the wastewater drainage system. Surface water from process areas is collected and diverted to the waste stream for treatment. They arise from production areas; boiler blows down and cleaning. This is also combined with domestic water which includes the

wastewater from the canteen and sanitary areas. The odour sources include the balance tank in the effluent treatment plant area and the final waste stream after treatment. Ambient air from these locations is collected and brought back to the laboratory for analysis.

Sampling

Samples are collected at two main odour source points identified in each plant: The balance tank (where dissolved air flocculation sludge dewatering takes place) and the final waste stream (after treatment). A sampling of ambient air from these locations is conducted using a standard dynamic sampling method which involves the use of a sampling pump to collect air samples over a specific period. The air samples are collected using sampling bags. Olfactometry is used to rate the intensity of the odour using a standardised scale. Based on the olfactometry procedure, a panel of qualified assessors performs an average of three measurements. The panellists are selected and trained using the criteria proposed for the dynamic olfactometry procedure (CEN 2003). Air samples are collected from the source and transported to the laboratory within 24 hours to determine odour concentrations. Samples are then diluted to a suitable concentration and introduced into an olfactometer. The panel evaluate the odour intensity, and the intensity is calculated based on the dilution ratio, the number of assessors and the average score assigned by the assessors. The results are reported in European odour units per cubic metre and include information on sample collection, preparation, and analysis procedures, as well as relevant information on the source of the odour (CEN 2003).

Model selection and input data

AERMOD atmospheric dispersion modelling software is used to predict the dispersion of odorous emissions in the atmosphere from wastewater streams. The model input data includes source characteristics, meteorological data, the topography of the area and odour emission rate (Tartakovsky *et al* 2013). Topographical data can be used in the models as it can significantly affect the odour plume dispersion and the predicted odour concentration at a specific odour-sensitive receptor. (Hayes *et al* 2006). The source characteristics include source type and dimensions, stack height, efflux velocity, temperature, and odour emission rates. Meteorological data include information like wind speed, wind direction, and temperature from the nearby weather stations to the seven processing plants that can be representative of typical climatological conditions of the processing plants.

Expected results

Effective management and treatment strategies can help minimise odour emissions from dairy wastewater. Air dispersion modelling will be used to predict the dispersion of odorous emissions in the atmosphere and evaluate the potential impacts on air quality and human health, which will inform the development of effective management and control strategies.

Conclusion

In conclusion, the investigation of odour emissions from dairy wastewater and their potential impact on the surrounding environment, especially the negative impacts on the health and well-being of both animals and humans living in the surrounding area, will aid decision-making at the plant level and the local authority in putting in place abatement and treatment systems to keep odour emissions within limits.

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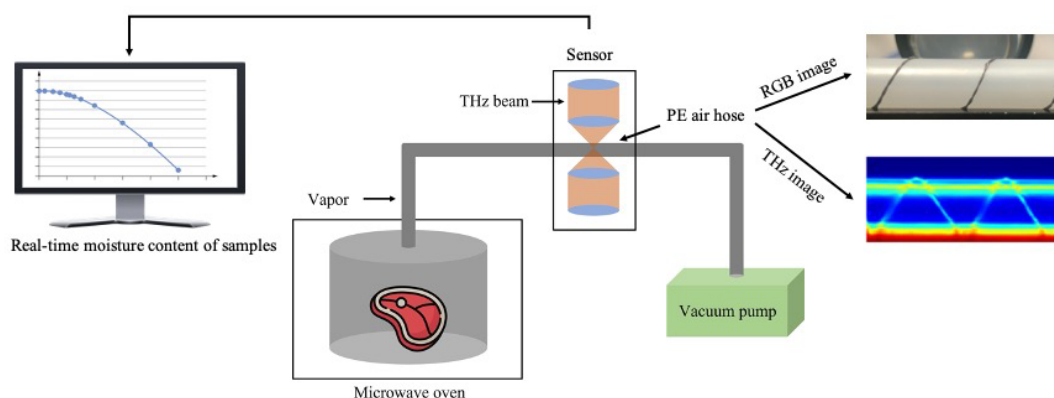
Yuqiao Ren, BE, M.EngSc.

Project Title: Advanced application of Terahertz time-domain spectral technique for in-situ monitoring of microwave vacuum dehydration

Project Leader: Prof. Da-Wen Sun

Abstract

Moisture content (MC) is an essential quality parameter in food drying processes; nevertheless, conducting non-destructive, in-situ analysis of dynamic MC in products throughout processing remains challenging. This investigation presents an in-situ, indirect measurement technique employing Terahertz time-domain spectroscopy (THz-TDS) to predict MC in real-time for food items during microwave vacuum drying (MVD). As MVD proceeds, THz-TDS persistently detects the evolving moisture vapour emanating from the desiccator via a polyethylene air conduit. The acquired THz spectra undergo processing to calibrate MC loss prediction models utilizing support vector regression, Gaussian process regression, and ensemble regression. Subsequently, MC is ascertained through moisture loss prediction outcomes. The most accurate real-time MC prediction results for beef and carrot slices achieved R^2 of 0.995, RMSE of 0.0162, and RDP of 22. The devised system introduces an innovative approach for examining drying kinetics during MVD and broadens the applicability of THz-TDS techniques within the food industry.



Selected Recent Publications

- Ren, Y., Lei, T., & Sun, D.-W. (2023). 'In-situ indirect measurements of real-time moisture contents during microwave vacuum drying of beef and carrot slices using terahertz time-domain spectroscopy', *Food Chemistry*, 418, 135943.
- Ren, Y., & Sun, D.-W. (2022). 'Monitoring of Moisture Contents and Rehydration Rates of Microwave Vacuum and Hot Air Dehydrated Beef Slices and Splits Using Hyperspectral Imaging', *Food Chemistry*, 132346.
- Ren, Y., Lin, X., Lei, T., & Sun, D.-W. (2021). 'Recent developments in vibrational spectral analyses for dynamically assessing and monitoring food dehydration processes', *Critical reviews in food science and nutrition*, 1-27.

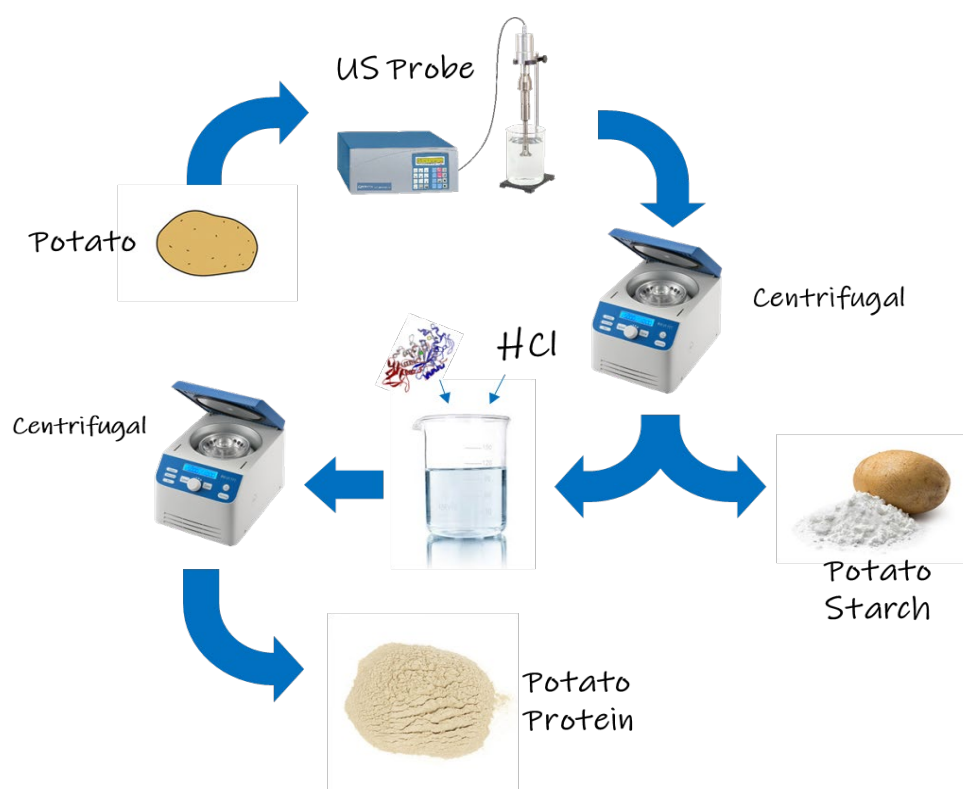
Zhipeng Hu, PhD

Project Title: Isolation of proteins from potato (*Solanum tuberosum*) using ultrasound and enzyme-assisted extraction techniques.

Project Leader: Da-Wen Sun, Brijesh K. Tiwari

Abstract

Potatoes are widely popular across the globe and are ranked as the fourth most grown crop in the world. The biological value of potato protein is 90, which is superior to other significant plant protein sources like legumes and soybean. Despite not being a typical protein source, potatoes' considerable yield and numerous beneficial by-products make them a valuable and potentially viable protein source. This study compared the extraction yield and energy consumption of some conventional and innovative methods for extracting potato protein and analyzed the extracts. The techniques used in the study are divided into two categories according to their solvent type: alkali and water. The methods consist of traditional methods, ultrasound-assisted extraction, two kinds of enzymes-assisted extractions, and a combination of ultrasound and enzymes-assisted extraction. In general, the yields of the alkali-based extraction methods were better than those of the water-based extraction methods. Among them, the alkali-based ultrasonic-assisted extraction obtained the highest protein yield (3.05g/100g), which was almost twice that of the conventional method (1.31g/100g and 1.57g/100g). Enzyme assistance had less impact on yield. Compared with the innovative methods, the two traditional methods obtained relatively higher extract purity (79.23% for water and 79.77% for alkali), which may be because the nova methods more effectively destroyed the cell structure and released more compounds. Ultrasonic-assisted extraction is undoubtedly the best choice in terms of energy consumption and processing time. It saves 57.7% of energy compared with conventional methods, and only needs 30 min, while conventional methods need 18 h.

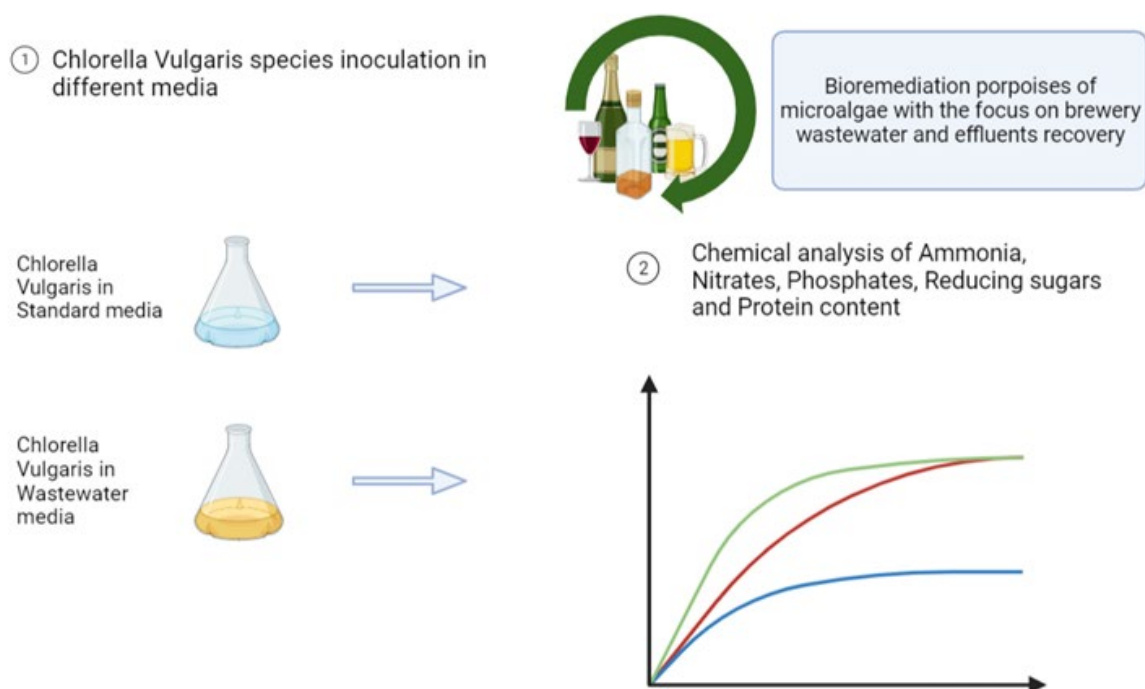


Project Title: Microalgae for the bioremediation and valorisation of brewery effluents

Project Leader: Dr. Ronald Halim

Abstract

Beer industry is a crucial pillar of the European economy, creating valuable products from simple ingredients such as barley, malt, water and yeasts. However, the production of beer requires massive amounts of water, most of which becomes waste once brewing processes are completed. Despite this, wastewater contains valuable nutrients such as glucose, proteins, amino acids, and other chemicals. Microalgae were used in this experiment to treat brewery wastewater, due to its capacity to remove pollutants from wastewater and effluents. It is associated with bioremediation procedures that can remove inorganic nutrients like nitrates, phosphates and ammonium while utilizing them to grow. The experiment aims to investigate the correlation of growth in different media, namely standard media and wastewater, and evaluate it through performing optical density measurements at specific wavelengths. Chemical analysis was also carried out to search for ammonia, phosphates, nitrates, reducing sugars and proteins. The preliminary evaluation indicated that microalgae exhibited persistent, exponential growth in wastewater media, implying a positive response to the aim of the study.



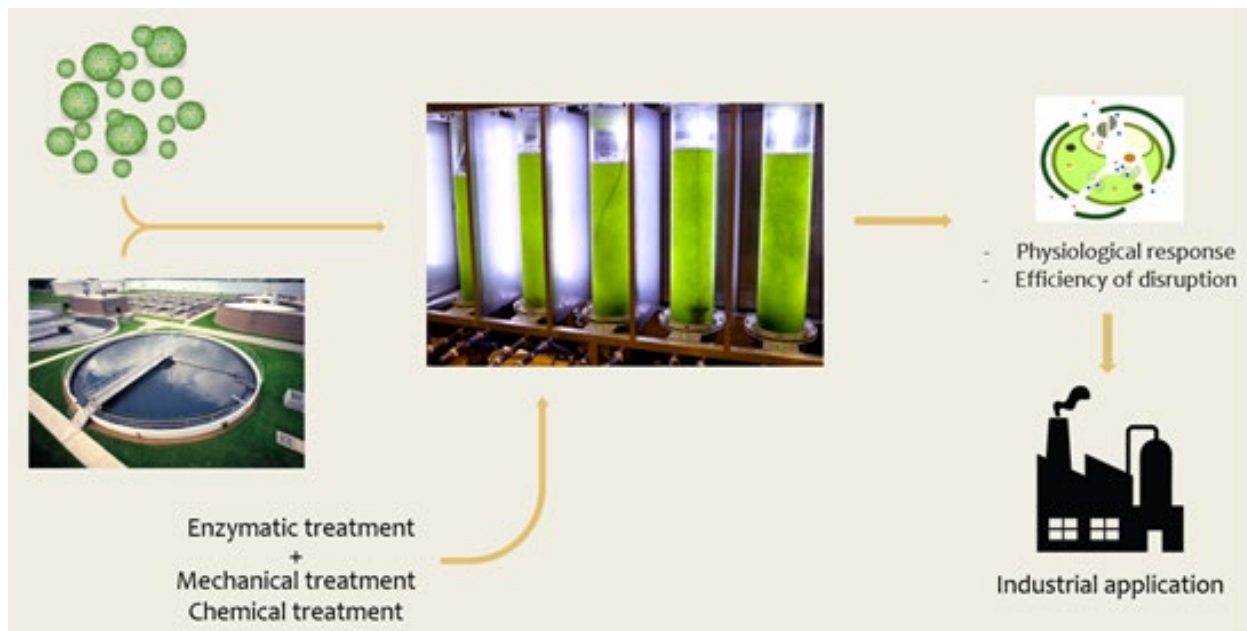
Nicholas Serlini, BSc, Msc.

Project Title: Understanding the ultrastructure of microalgae cell walls to enable biomass fractionation

Project Leader: Dr. Ronald Halim

Abstract

For modern society, microalgae represent one of the most promising options for the production of metabolites to cope with increasing demands in food, biochemical and energy sectors. As novel organisms though, microalgae often present issues related to the lack of knowledge possessed by researchers, leading to less efficient approaches and techniques for their exploitation. This is also reflected by the few processes that have been used so far in industry. One of the most immediate problems concerns microalgae cell structures, specifically the cell wall architecture, which serve as barrier between these species and extracellular environment, also preventing cost-effective recovery of intracellular products from the biomass. Our project will focus on analysing the cell wall of different microalgae, with particular interest on the rich-lipid species *Nannochloropsis oceanica*, to understand its composition and organisation. The goal will be to couple these results with knowledge regarding the optimization of enzymatic treatments, obtaining cell wall disruption methods that could be energy-efficient, less environmentally harmful and non-degradative towards the biomass (thus preserving product quality) that will have to be extracted.



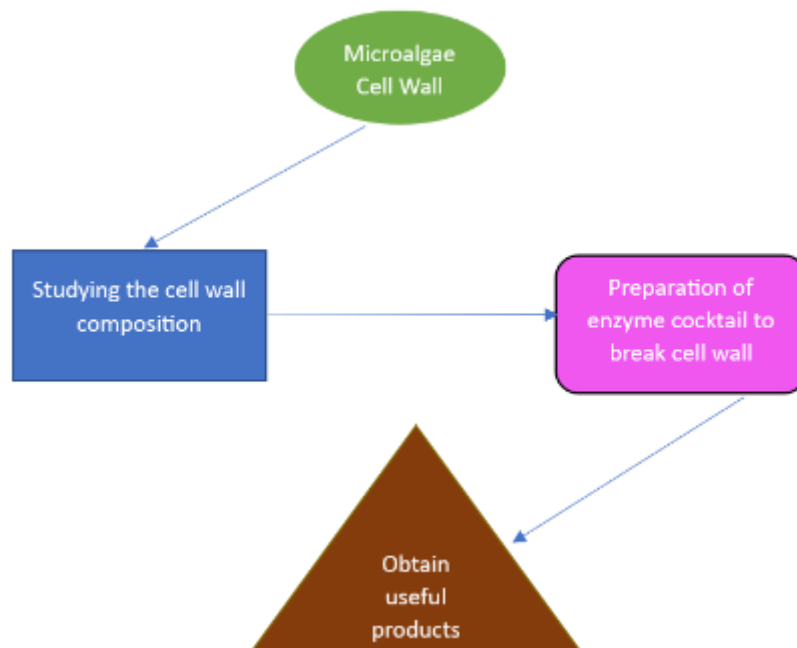
Sneha Shivakumar, BSc, MEngSc.

Project Title: Microalgae cell-wall analysis and enzymatic disruption

Project Leader: Dr. Ronald Halim

Abstract

Some of the major global issues at present are reduction of carbon dioxide emissions, bioremediation of sewage, and production of sustainable biofuels and enhancement of nutrition in human and livestock diet. Research has proven that microalgae can be a key player to combat these issues as they can produce valuable products like lipids and proteins. Previous research has shown that most of the microalgal products are intracellular and can be obtained only after being liberated from the cell wall encapsulation. The multilayered and rigid cell walls found around the microalgae cells of some genera of microalgae provide them with protection against the external environment but has become a significant problem in the path for the commercialization of microalgae products. The present mechanical and chemical methods have several drawbacks. This project aims to overcome this obstacle by conducting a thorough investigation on the cell walls of *Nannochloropsis* species and *Chlorella* species. The study will then prepare an enzyme cocktail specific to break the cell walls of the microalgae, hence facilitating upscaling of a more commercially viable microalgae system.

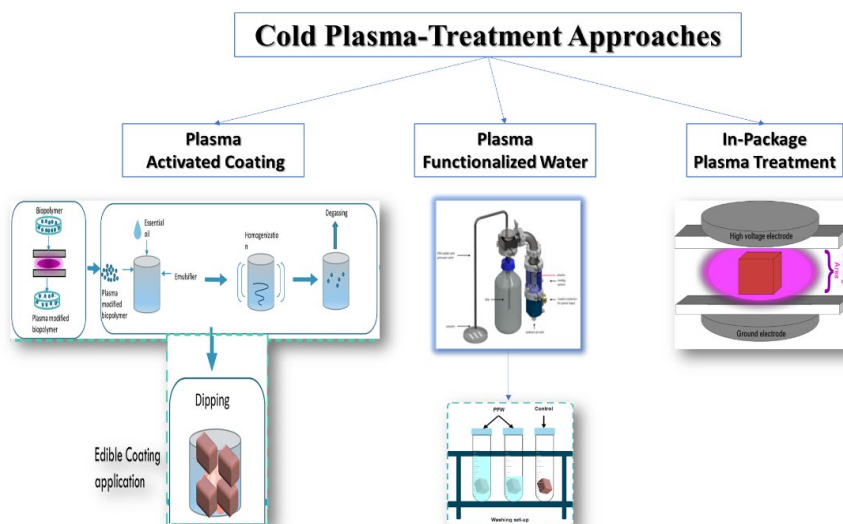


Project Title: Chicken Juice a food-based Model to Assess the antimicrobial efficiency of three Cold plasma approaches

Project Leader: Professor Paula Bourke

Abstract

Cold plasma (CP) is a cutting-edge technology flexibly applied to control food-related contaminations. The present study suggested a framework of CP interventions to control pathogens in the poultry-processing chain. Individual and combined effects of Plasma Functionalized Water (PFW), In-package CP, and active chitosan coating encapsulating plant essential oils (lemongrass, oregano, and thyme) were investigated. In-package DBD and MidiPlex were the systems considered. Their bactericidal effects identified against suspended planktonic cells and attached biofilms in a sterilized chicken-juice model (liquid or solid) to mimic the poultry-processing environment. CP efficiency tested against *Salmonella* Typhimurium and *Campylobacter jejuni* regarding plasma process parameters. Within 15-sec of PFW treatment, 9.26 and 6.08 log₁₀CFU/ml reduction reached of *S. Typhimurium* and *C. jejuni* respectively. In-package treatment (3-min) reduced *S. Typhimurium* and *C. jejuni* by 2.9 and 3.3 log₁₀CFU/ml respectively. Active-coating with lemongrass or thyme reduced bacterial load by 2.03 log₁₀CFU/ml. Where plasma-functionalized-chitosan coating displayed 1.59 log₁₀CFU/ml inactivation. Combining the optimum treatments reduced pathogen levels by approximately 5-log cycles. CP approaches are effective, scalable, and sustainable. They can be applied at individual or sequential process intervention points to rapidly control poultry-related pathogens present in suspension, attached in biofilms to abiotic surfaces or on the surface of poultry meat.



Selected Recent Publications

- Barroug, Soukaina, Sonal Chaple, and Paula Bourke. 2021. "Combination of Natural Compounds With Novel Non-Thermal Technologies for Poultry Products: A Review." *Frontiers in Nutrition* 8 (June): 1–15. <https://doi.org/10.3389/fnut.2021.628723>.
- Marzlan, Anis Asyila, Anis Shobirin, Meor Hussin, Paula Bourke, Sonal Chaple, Soukaina Barroug, and Belal J Muhialdin. 2021. "Combination of Green Extraction Techniques and Essential Oils to Develop Active Packaging for Improving the Quality and Shelf Life for Chicken Meat Combination of Green Extraction Techniques and Essential Oils to Develop Active Packaging for Improving The." *Food Reviews International* 00 (00): 1–23. <https://doi.org/10.1080/87559129.2021.2013499>.

INVESTIGATING THE DEHYDRATION FEATURE OF BEEF PRODUCTS WITH DIFFERENT PRE-TREATMENT DURING MICROWAVE VACUUM DEHYDRATION USING NIR-HSI AND TERAHERTZ SPECTRAL ANALYSES

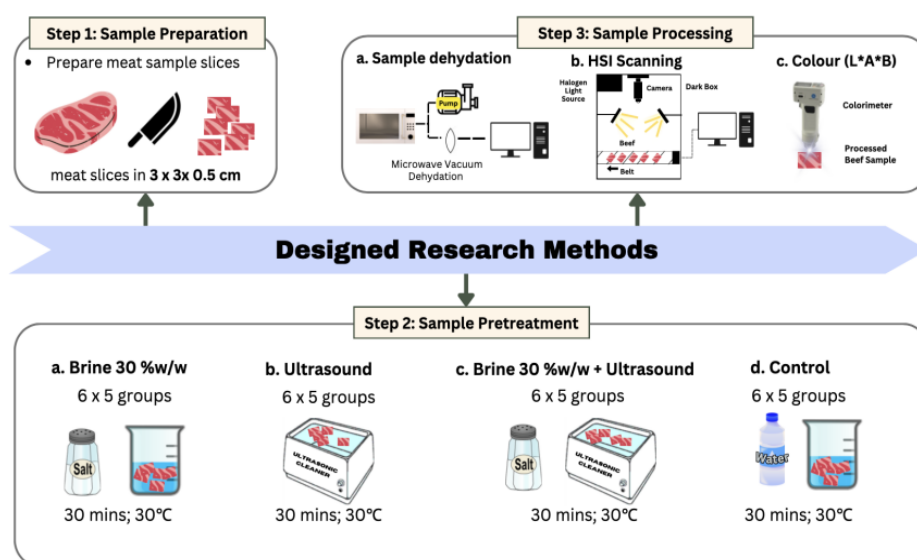
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Abstract

With the rising concern of food quality control, a series of pre-treatment and detecting methods are implemented to improve the accuracy and efficiency. The utilisation of non-thermal drying pre-treatment methods can improve the unpredictable variations in product quality due to non-uniformity in the microwave vacuum dehydration (MVD) process and thus enables the improvement of the quality of the dried product. In this study, four different non-thermal pretreatment methods including ultrasonic treatment, osmotic dehydration, ultrasonic-assisted osmotic dehydration and control, were used to investigate the effect of different non-thermal pretreatment methods on the quality of dehydrated beef. The results show that the use of osmotic drying pretreatment significantly improved the drying efficiency of MVD, while reducing the variation of brightness, total colour difference and shrinkage. In addition, the moisture distribution obtained from NIR-HSI can be observed, and the role of ultrasonic pretreatment in reducing drying inhomogeneities was found, while osmotic pretreatment may aggravate the inhomogeneous distribution.

Graphical abstract



Introduction

Beef, as one of the primary components in the public's diet, contains numerous macro- and micronutrients including proteins, vitamins and fibre that contribute to maintaining the healthy status of human bodies. With the expanding population, the demand of beef increases, thus the quality of beef has become a key parameter for the industry to sustain and expand the market. Food quality assurance of such products has been brought to the discussion due to their high depletion rates based on the intricate structure of meat and diverse processing technologies (Damez and Clerjon, 2013).

Furthermore, meat is putrescible under inappropriate storage conditions which sharpens the issue of quality assurance (Kutsanedzie *et al* 2019).

With the rapid development of the food industry, an increasing manufacturing efficiency led by the advanced global manufacturing and management system can be observed, which results in a rising concern regarding the issue of food product quality has been shown by the public. However, the majority of traditional quality examination methods are considered to be time consuming, labour intensive, destructive and costly. Therefore, a rapid, non-destructive and economical method is required to adapt to the current circumstance. Terahertz (THz) spectroscopy has been considered as one of the non-ionizing technologies that are desired for such activity. Its working principle is based on the detection of THz radiation penetrated through the materials, thus determining the required food properties. The THz radiation consists of waves that fall within the terahertz region which refers to a narrow gap between microwave and infrared regions of the electromagnetic spectrum with a typical wave range of 0.3 to 30 THz (Abbasi *et al* 2023). The application of THz spectroscopy has been implemented in various fields in the food industry including the moisture content determination (Afsah-Hejri *et al* 2019).

The objective of this thesis is to investigate the impact of different pretreatment methods (i.e. brine, ultrasound, brine and ultrasound and osmotic dehydration) on the dehydration feature of beef during the microwave dehydration process by using the NIR-HSI and THz spectral analyses.

Methods

Sample preparation

Beef round cuts were purchased from the local supermarket, connective tissues and fat on the outer surface of the beef round cuts were removed and the cuts were frozen after processing to ensure the consistency for slicing. The frozen beef cuts were sliced with the thickness of 4mm by the slicer transversely and longitudinally. A sharp-edged knife was used to chop the meat slices into a square shape of 30mm x 30mm. A total of 240 samples were prepared with allocation of five samples per group. Assigned groups were sealed in plastic petri dishes and stored under refrigeration at 4°C to minimise the impact of dehydration and oxidation.

Drying pretreatment

Four different drying pretreatments of ultrasound pretreatment (i.e. US, distilled water, 30°C, 60 mins), osmotic dehydration (i.e. OD, 10% w/v brine, 30°C, 60 mins), ultrasound-assisted osmotic dehydration (i.e. UOD, 10% w/v brine in ultrasound bath, 30°C, 60 mins), and control (i.e. distilled water, 30°C, 60 mins) were applied to the practical. To improve the accuracy, duplicate experiments of each method (i.e., $4 \times 2 = 8$ groups in total) were employed. Excessive water remaining on the beef samples were blotted by tissues.

Drying process

Microwave vacuum drying (MVD) technique was implemented in this study. The drying process used is similar to that used by Ren and Sun (2022).

Hyperspectral images acquisition

Pretreated samples were scanned by using the NIR-HSI system where samples were placed on the moving belt at 20 mm/s with exposure time of 10 ms per shot, in order to achieve the line-scanning mode for image acquisition. Then the enhancement of raw images were performed by reflectance adjustment aiming at minimising the noise generated by light source variation. After the separation of regions of interest (ROI) from the background, the Otsu thresholding method, L1HyMixDe algorithm, and the MATLAB function of regionprops were implemented while analysing the images and their pixel numbers to remove the background noise and improve the accuracy of sample order based on the mass centre of each sample. Thus, the separation of images and calculation of average spectra were achieved.

Analysis of reference moisture

AOAC method was implemented to analyse the reference moisture content (G. J. E. von Gersdorff *et al* 2021). The equation presented below (was implemented for the calculation of moisture content.

$$MC (\%) = \frac{m_t - m_d}{m_t} \times 100$$

where MC (%) is the moisture content, m_t represents the sample after t min MVD process; m_d represents the dry matter weight of the sample after 24 hour oven drying.

Terahertz spectroscopic data processing

The fast Fourier transform was used to convert the obtained THz time-domain spectra to frequency domain spectra. The calculation of absorption coefficient differences at determined intervals of the dehydration stage were determined depending on the THz spectra data of air background and PE pipe. The trapezoidal numerical integration method was implemented during the computation of numerical integration of THz absorption coefficients.

Models calibration and evaluation

Since two different methods were applied to the MC prediction regarding the direct and indirect sensing of real-time MC loss using NIR-HSI and THz-TDS respectively, the Partial Least Squares Regression (PLSR) method was used, aiming at comparing the prediction accuracy of two methods. The equation presented below illustrates the calculation of real-time MC.

$$MC = MC_i - MC_{loss \cdot t}$$

where MC_i is the reference MC of fresh sample, and the $MC_{loss \cdot t}$ is the predicted moisture loss at t mins during processing (Ren and Sun, 2022).

Two key elements of R^2 and RMSE were determined and applied to support the accuracy investigation of calibration and validation. All the collected data were processed and analysed by using MATLAB 2021b (The MathWorks, Inc., Natick, MA, USA). The equations of calculating R^2 and RMSE are listed below.

$$R^2 = 1 - \frac{\sum_{i=1}^n (\hat{y}_i - y_i)^2}{\sum_{i=1}^n (\hat{y}_i - \bar{y})^2} \quad RMSE = \sqrt{\frac{1}{n} \sum_{i=1}^n (\hat{y}_i - y_i)^2}$$

where \hat{y} is the predicted values, y is the measured values, \bar{y} is the average of samples, and n is the number of sample spectra (Ren and Sun, 2022).

Results and discussion

Drying kinetics of samples under different pretreatment

An average initial moisture content (MC) of 77.99% with a standard deviation of 1.456 was obtained among four groups. A continuous decline in MC was observed among all the groups during the dehydration process. A range of mean final MC between 7.51% to 6.15% was discovered which proposes the dependence of dehydration speed with different methods of pre-treatment. Moreover, the utilisation of brine in pre-treatments (i.e., OD and UAOD) exhibited a lower MC after 12 minutes of the commencement of the dehydration process than samples in other two groups.

Spectral analysis and prediction results

Among the results, the mean reflectance spectra of four groups exhibits an increasing trend with the processing of MVD. Their characteristics consist of similar curves but different amplitudes which can be caused by the decreasing MC in samples during dehydration and resulted in a lower absorbance of NIR radiation, thus leading to a high spectral reflectance (Liu *et al* 2018). Moreover, this phenomenon is more significant among the groups where the ultrasound was implemented (i.e., UL and UAOD), that suggests the role of ultrasound in promoting the reflectance of samples.

Furthermore, the real-time MC and MC loss prediction can be generated via the spectral data recorded from THz-TDS and NIR-HSI, which satisfy the prediction results for both terms. However, a better MC loss prediction was performed by the THz-TDS in-direct sensing method with R_p^2 of 0.9817 and RMSE_p values of 0.0112.

Conclusion

To conclude, this thesis mainly focuses on the investigation of the impact of different pretreatment methods (i.e., osmotic drying, ultrasound-assisted osmotic drying and ultrasound) on the dehydration characteristics of beef samples during microwave vacuum drying by using NIR-HSI and Terahertz spectral imaging. The osmotic drying as the pretreatment enables the maximum enhancement of dehydration speed. Moreover, the utilisation of ultrasound is able to inhibit the non-uniform moisture distribution in beef samples during MVD processing. Additionally, the higher performance of THz-TDS on aspects of higher accuracy of prediction results was demonstrated.

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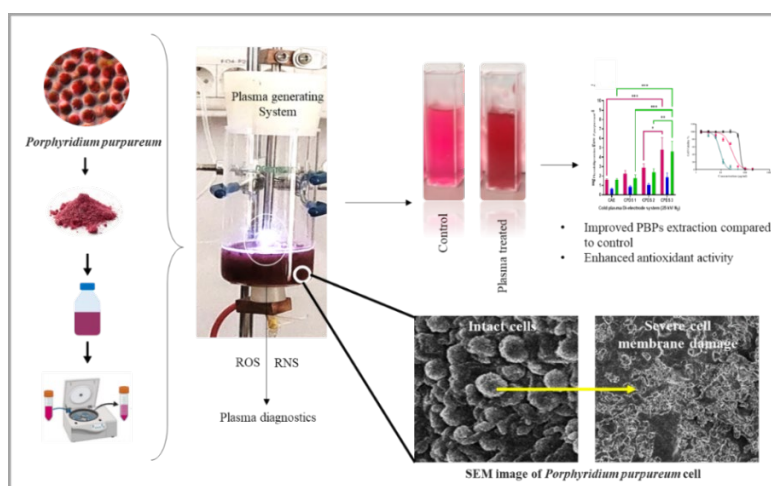
Shaba Noore, PhD

Project Title: Extraction yield and biological activity of phycobiliproteins from *Porphyridium purpureum* using atmospheric cold plasma discharge and jet systems

Project Leader: Prof. Colm O'Donnell

Abstract

Phycobiliproteins (PBPs) extracted from *Porphyridium purpureum* (*P.p*) have bioactive properties and are widely used as ingredients in nutraceutical and food applications. This study investigates the use of two cold plasma systems, namely the cold plasma discharge system (CPDS) and the cold plasma jet system (CPJS), for the aqueous extraction of PBPs from *P.p*. The extraction yield of PBPs is calculated using UV-Vis spectroscopy over a spectral range of 250-700 nm. Three types of PBPs, namely phycoerythrins (PEs), phycocyanins (PCs), and allophycocyanins (APCs), are identified in the crude extracts obtained. CPDS treatments are shown to be more effective than CPJS treatments in increasing extraction yield. The highest PBPs extraction yield (11.31 ± 1.02 mg/g DW *P.p*) is obtained from CPDS-treated samples at 25 kV using N₂ for 9 min. CPDS treatments are also shown to be more effective than CPJS treatments in increasing the antioxidant activities of the PBPs crude extracts obtained. PBPs obtained using CPDS (25 kV; 9 min; N₂) have the highest DPPH (IC₅₀ of 9.0 ± 0.75 μ g/mL) and FRAP (207.34 ± 12.96 μ mol/L) antioxidant activities observed. Cytotoxicity analysis shows the growth inhibitory potential of the PBPs obtained from plasma-treated samples in Caco-2 human colorectal adenocarcinoma cell lines. PBPs obtained from samples treated with CPDS (25 kV; 9 min; N₂) exhibit the highest cytotoxicity potential in Caco-2 human colorectal adenocarcinoma cell lines. This study demonstrates that cold plasma treatments increase the extraction yield of PBPs obtained from *P.p* and also enhance the antioxidant and cytotoxic properties of the PBP crude extracts.



Selected Recent Publications

Noore, S.; Joshi, A.; Kumari, B.; Zhao, M.; O'Donnell, C.; Tiwari, B.K. (2022) Effects of novel extraction strategies on the recovery of phenolic compounds and associated antioxidant properties from buckwheat hull (*Fagopyrum esculentum*). *Processes*, 10, 365.

GROWTH OF LISTERIA MONOCYTOGENES IN MOZZARELLA CHEESE

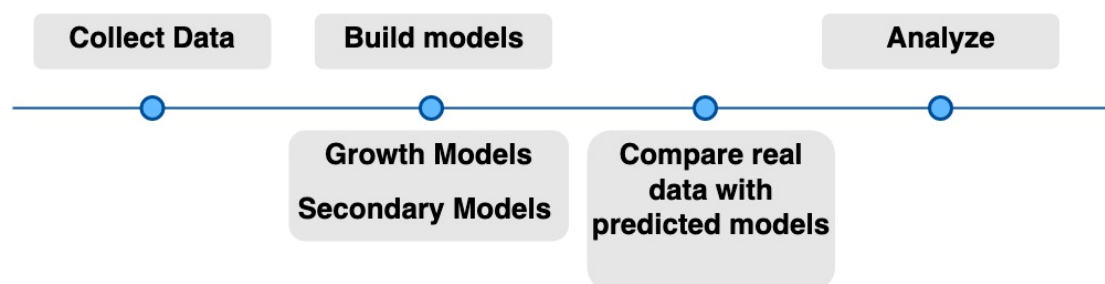
Chuqin Sun, Francis Butler

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

With food safety becoming increasingly important worldwide, *Listeria monocytogenes* is ranked one of the deadliest foodborne pathogens due to its considerable pathogenicity. Mozzarella cheese is a type of soft cheese, its high-water content provides a suitable environment for the growth of *Listeria monocytogenes*, and traditional cooking methods are not sufficient to destroy the potential *Listeria monocytogenes* in cheese, therefore, it is essential to model the growth of the pathogenic bacteria throughout the storage process for the shelf life of the product as well as to extend its shelf life. In this paper, the primary and secondary models of *Listeria monocytogenes* growth in mozzarella cheese were constructed based on data from databases and literature, respectively, and the critical factor (temperature) affecting the growth of *Listeria monocytogenes* in mozzarella cheese was analyzed. After comparing the fitting results of Gompertz and Baranyi with the actual data points, it was confirmed that the Gompertz model was more suitable for the growth of *Listeria monocytogenes* in mozzarella cheese.

Graphical Abstract



Introduction

Listeria monocytogenes can cause listeriosis in humans which is a severe foodborne disease. *Listeria monocytogenes* can adapt to a variety of environments and even extreme conditions, reproducing normally at 4°C, surviving for up to a year at -20°C, and growing at a low water activity (<0.94), low pH (<4.4), and high salt concentration (10% NaCl) (Rückerl *et al* 2014). One of the items with the highest rate of *Listeria monocytogenes* contamination is cheese. Semi-hard or hard cheeses seem less prone to contamination than soft-ripened cheeses. Combining the organism's tolerance to cold and salt, and the fact that soft cheeses are typically served without any sterilization (cooking, heating) to remove contaminants introduced during the entire production and ripening period, soft cheeses offer favorable conditions for the growth of *Listeria monocytogenes*.

The *Listeria monocytogenes* growth is also greatly supported by the considerable pH increase that occurs during the surface maturation of certain dairy products (Guenther and Loessner, 2011). In 1985, many pregnant women and children in the United States suffered from vomiting, shortness of breath, and miscarriage after eating soft cheese, which eventually led to a tragic accident in which 142 people were poisoned, and 52 died; the investigation found that the cheese was contaminated with the foodborne pathogen *Listeria monocytogenes* (Yoon *et al* 2016).

Mozzarella is a soft, unripe pasta filata (spun paste) cheese made from pasteurized or unpasteurized milk, often made with cow's milk or buffalo milk, which can provide suitable conditions for the growth of *Listeria monocytogenes* (Tirloni *et al* 2019). As the storage time increases, the pH becomes higher and more ideal for developing *Listeria monocytogenes*.

This study aims to find the most suitable model by comparing the actual data with the prediction model through modeling, analyze the effect of microorganisms in cheese on the growth of *Listeria monocytogenes* and the main influencing factors such as temperature, pH and water activity, and then discuss practical ways to extend shelf life.

Materials and Methods

Data collection

Data should be extracted from literature, database, and the government's official website, some data were obtained from tables and graphs in the literature, and graphs were digitized using MATLAB.

Develop growth survival models

Extracted data were used to build growth survival models and secondary growth models and optimize models. Fit multiple sets of data from different sources and compare different models (Baranyi and Roberts model, and Gompertz), and determine the best one. where the Baranyi and Roberts model is fitted with online DMFit on combase.

The equation of Gompertz is: $LgN_t = LgN_0 + C * e^{-e^{\left(\frac{-B}{1+e^{\left(\frac{t}{M-\frac{t}{\alpha}}\right)}}\right)}}$

As shown in the equation, N_0 and N_t are the numbers of bacteria at the moment t and the number of bacteria at the moment of starting 0, respectively; C is the difference between the number of microorganisms entering the stable phase and the initial value; B is the maximum growth rate in the logarithmic phase; M is the moment corresponding to the maximum growth rate in the logarithmic phase.

The most suitable model is determined by fitting the obtained data in two different models (Gompertz and Baranyi), according to the value of R-square and RMSE.

Develop secondary growth models

After modeling the environmental factors including temperature, pH, and water activity, and determining the key factor. Compare different secondary models including Response Surface Equation, Square root model, and Arrhenius model, and determine the most suitable one.

Expected Results and Discussion

The data points were from the pieces of literature and some of them were excerpted by using Grabit in MATLAB, and then used in Gompertz and Baranyi models, respectively. Taking Han et al. (2014) as an example, the experimental data were substituted into the two models to obtain different fitted curves, the concentration of *Listeria monocytogenes* over time is shown in Figure 1 (a). Based on the R-Square value, Gompertz 0.988 > Baranyi 0.964, and the RMSE value Gompertz 0.206 < Baranyi 0.278, after the accuracy of the model is verified, it can be concluded that the Gompertz model is more suitable for predicting the growth of *L. monocytogenes* in this data set.

The number of *L. monocytogenes* corresponding to concentration at different temperatures (the main influencing factor expected) was recorded using the Gompertz model. The estimated exponential growth rate of *L. monocytogenes* on mozzarella cheese is shown in Figure 1(b).

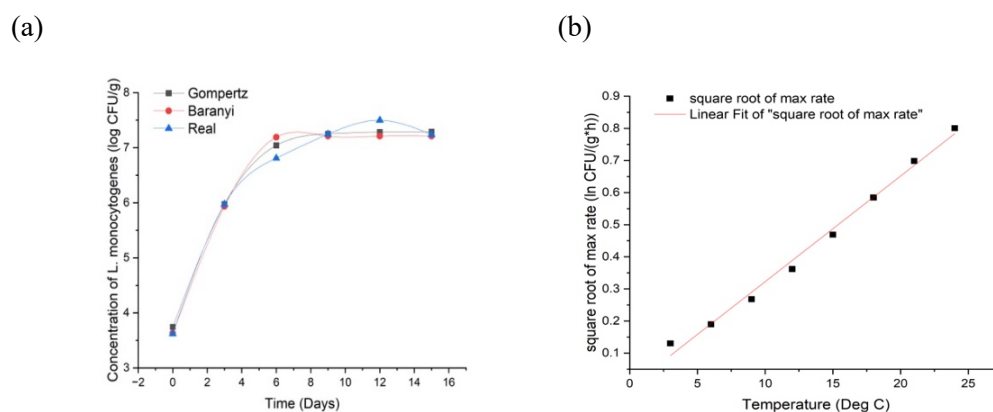


Figure 1. (a) Comparisons among Gompertz, Baranyi and Roberts model and real points and (b) Estimated square-root model for the growth of *L. monocytogenes*.

Conclusion

In this paper, primary and secondary models were constructed for the growth of *L. monocytogenes* in mozzarella cheese with and without pasteurization treatment based on data from databases and literature, respectively, to analyze the key factors affecting the growth of *Listeria monocytogenes* in mozzarella cheese. By comparing the degree of conformity of the Baranyi model and Gompertz model with the experimental data points, combined with the quantitative analysis of R^2 and RMSE, the primary model that best met the conditions of this paper was determined to be the Gompertz model. It is expected that the secondary growth model constructed based on the primary model yields an effective temperature range that best suppresses the growth of mozzarella. Subsequent experiments can also be conducted to verify the validity of the model and further optimize the model.

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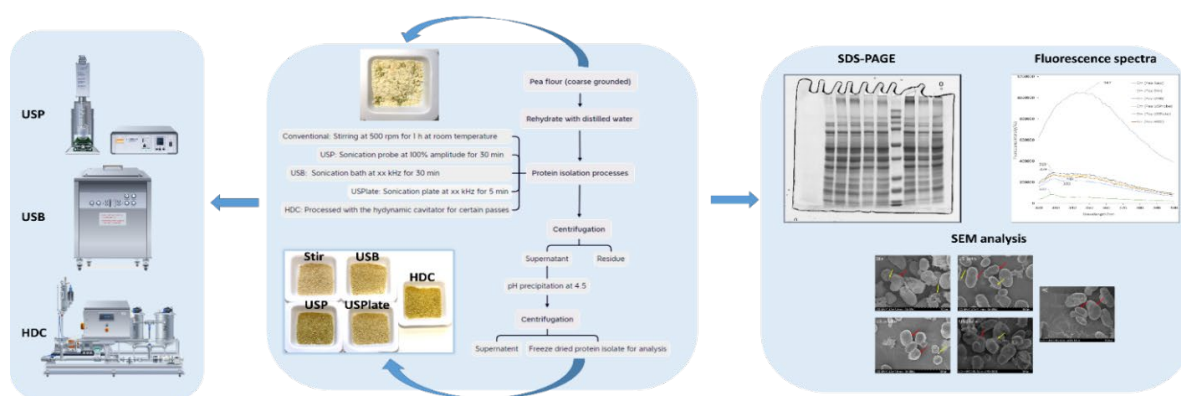
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Project Title: Influence of cavitation technologies, mainly hydrodynamic and ultrasonic cavitation, on structure and functional properties of pea protein

Project Leader: Prof. Da-Wen Sun and Prof. Brijesh K Tiwari

Abstract

Pea protein has become a popular alternative protein source in various food products due to its nutritional value. With the increasing interest in plant protein extraction using advanced technologies, cavitation-based processing has been extensively studied due to its low energy consumption and high processing efficiency. The cavitation phenomenon, which releases high energy by generating and collapsing bubbles, has proven to be effective in enhancing food processing efficiency (Tang et al., 2023). The present study aims to investigate the influence of different cavitation technologies on the structure and functional properties of pea protein. To carry out the study, 50 g of pea flour was dispersed in 1000 mL of distilled water at a ratio of 1:20 w/v, and the pH was adjusted to 10 using 1M NaOH. Then different cavitation technologies (ultrasound probe, ultrasound bath, ultrasound plate and hydrodynamic) as well as conventional stirring were used to process the sample. Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) and liquid chromatography coupled with electrospray-ionization quadrupole time-of-flight mass spectrometry (LC-ESI-QTOF/MS) were used to assess protein profile. Fluorescence analysis, scanning electron microscopy (SEM) and functional properties analysis were used to assess protein structure and functional properties. The hydrodynamic treatment demonstrated the highest protein content, with values of up to $80.35 \pm 2.18\%$, and all cavitation technologies exhibited higher protein content than conventional stirring. While stirring resulted in insufficiently broken granules, the SEM results revealed that the cavitation techniques could relatively completely disentangle the starch granules embedded in the cell matrix, illustrating that cavitation could enhance the separation of intracellular granules to enhance protein extraction due to the cavitation effects. With regard to functional properties, a significant difference in turbidity and total colour difference was observed between hydrodynamic and conventional treatments. This study found that cavitation can influence the protein profile and improve protein properties to some extent.



Selected Recent Publications

Tang, J., Zhu, X., Jambrak, A. R., Sun, D.-W., & Tiwari, B. K. (2023) ‘Mechanistic and synergistic aspects of ultrasonics and hydrodynamic cavitation for food processing’, *Critical Reviews in Food Science and Nutrition*, 1-22.

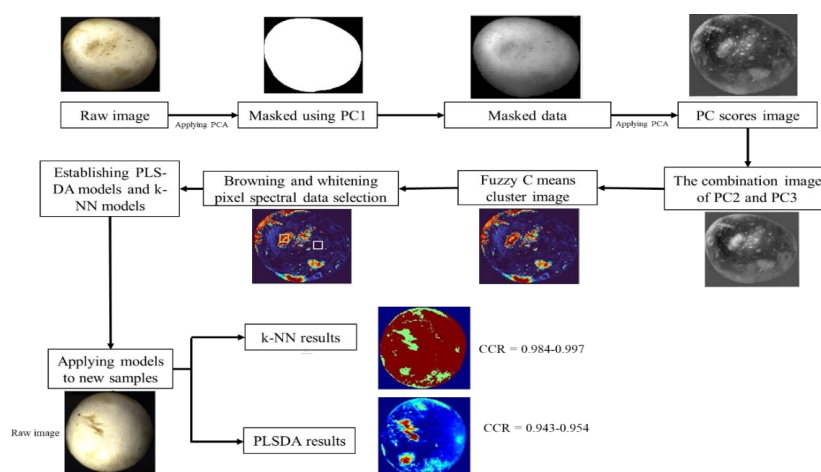
Kai Yang, MSc

Project title: Identification of mushroom browning patterns using a portable hyperspectral imaging camera combined with machine learning techniques

Project Leader: Dr Dimitrios Argyropoulos

Abstract

The application of hyperspectral imaging (HSI) technology as a non-destructive and rapid analytical tool for the detection of browning effects on white button mushrooms (*Agaricus bisporus*) has been under investigation since 2008. However, this technology is still in the experimental phase and has not yet been widely adopted for practical use. To address this, the present study aimed to evaluate the potential of a portable hyperspectral imaging camera operating in the visible-near infrared (400-1000 nm) wavelength range for the discrimination of browning and non-browning tissues in mushrooms. A combination of unsupervised training algorithms, namely Principles Component Analysis and Fuzzy C-means Clustering was employed for mushroom image segmentation and reference data selection. Subsequently, supervised classification models of k-nearest neighbor (KNN) and partial least square-discriminant analysis (PLS-DA) were developed to classify browning and non-browning mushroom tissues based on the reference data selected. The KNN and PLS-DA classification models exhibited corrected classification ratios (CCR) of 0.984-0.997 and 0.943-0.954, respectively. In summary, this study demonstrated the potential of a portable hyperspectral imaging camera in combination with machine learning models for the purpose of post-harvest mushroom quality control. The findings have important implications for the agri-food industry, particularly with regard to enhancing the efficiency and accuracy of quality control processes for mushrooms and other produce.



Selected Recent Publications

Yang, K., Zhao, M. and Argyropoulos, D., 2022. Detection of mushroom browning using RGB image segmentation approaches combined with hyperspectral image analysis. In 36th EFFoST International Conference 2022, November 7-9, Dublin, Ireland.

RISK ASSESSMENT OF *LISTERIA MONOCYTOGENES* IN BUTTER

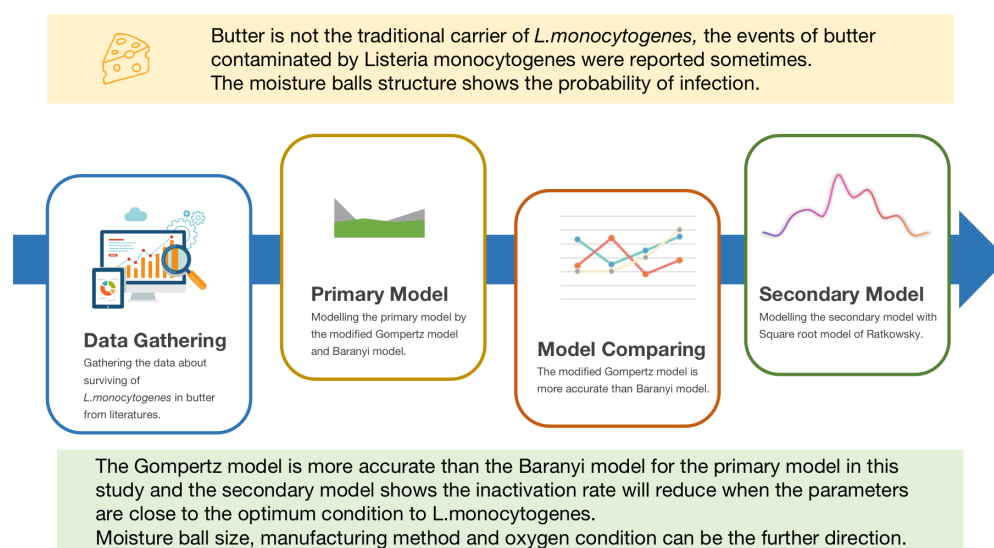
Zongyi Zhang, Francis Butler

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Abstract

Butter is not considered as the carrier of *Listeria monocytogenes* traditionally, however, the events of butter contaminated by *Listeria monocytogenes* were reported sometimes. The moisture balls structure shows the probability of infection which increases the risk of butter products. This study established models about surviving of *L.monocytogenes* in butter with different parameters including temperature, pH, water activity and salt content. The modified Gompertz model, Baranyi model were used for the primary model and the Square root model of Ratkowsky was used for the secondary model. The Gompertz model is more accurate than the Baranyi model for the primary model in this study and the results of the secondary model shows the inactivation rate will reduce when the parameters are close to the optimum condition to *L.monocytogenes*. As there are some other influence factors including moisture ball size, manufacturing method and oxygen condition can also affect the surviving of *L.monocytogenes* efficiently, further study can focus on these factors.

Graphical Abstract



Introduction

Butter is not considered as the traditional carrier for *listeria monocytogenes* because of its high fat content. However, there are still reports about the *listeria monocytogenes* found in butter frequently around the world. Finland first reported the outbreak of listeriosis in 1999 including 20 patients and 6 death cases which is also the first time *Listeria monocytogenes* has been isolated from butter (Lyytikäinen *et al* 2000). Later, *listeria* contamination of butter was often reported all over the world (Lewis *et al* 2006).

The microstructure of butter is many small moisture balls spread in the fat (Fig.1). *Listeria monocytogenes* is difficult to grow in the fat content and mainly remains in the moisture balls with nutrients including water and milk solids (Goff *et al* 1996). *Listeria* can survive in the moisture balls for a long period and cause listeriosis. As the long period of *Listeria* survives in butter, the risk of butter is growing.

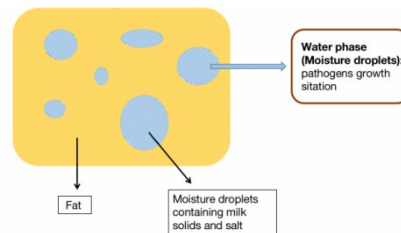


Figure 1. The microstructure of butter.

The exposed pathways of *L.monocytogenes* in butter are existing in the complete manufacturing line and consumer preservation stage. Establishing the survival model of *Listeria* in butter is efficient to analyse the *Listeria* level in butter and useful for the exposure assessment in risk assessment of *L.monocytogenes* in butter.

The objective of this study is to establish a survival model of *Listeria monocytogenes* in butter by models named logistic, modified Gompertz, Baranyi and Square root model of Ratkowsky.

Materials and Methods

Data gathering

Gathering data of surviving *Listeria monocytogenes* in butter with the parameters of pH, temperature, water activity and salt concentration separately from literature.

Primary Modelling

Fitting time versus log number of *Listeria monocytogenes* in butter and calculating the maximum surviving rates in different temperature, pH, water activity and salt concentration with the modified Gompertz and Baranyi model (Table 1). Comparing the two models with the real survival curve.

Second modelling

Modelling the maximum surviving rate versus different parameters respectively with the Square root model of Ratkowsky (Table 1).

Table 1. Models used in modelling the survival of *L.monocytogenes* in butter

Model type	Model name	Source
Primary Model	Modified Gompertz	(Bhaduri <i>et al</i> 1991)
	Baranyi	(Baranyi and Roberts 1994)
Secondary model	Square root model of Ratkowsky	(Ratkowsky <i>et al</i> 2005)

Results

Prediction of survival model with pH

The survival curves of *L.monocytogenes* in butter are fitted by Matlab with the model functions. The results by the modified Gompertz model are very similar to the actual curves (Fig 2) in which the R^2 of every curve is more than 0.99. Conversely, the Baranyi model has a more inaccurate result. Therefore, the modified Gompertz model is more suitable for establishing the pH model in this study. The maximum inactivation rates of bacteria are 1.269, 5.13, 0.5292, 0.8658, 5.616, 0.6852 at pH 4.05, 4.51, 4.58, 5.34, 5.38, 6.40 respectively. The rate at pH 4.51 and 5.38 are very different with other data which may be caused by the type of butter. As the pH increases, which is also closer to the optimum pH of *L.monocytogenes* at 6.5 to 7.0 (Cheng *et al* 2015), the trend of inactivation rate is reduced. And this trend will also fit for the secondary model.

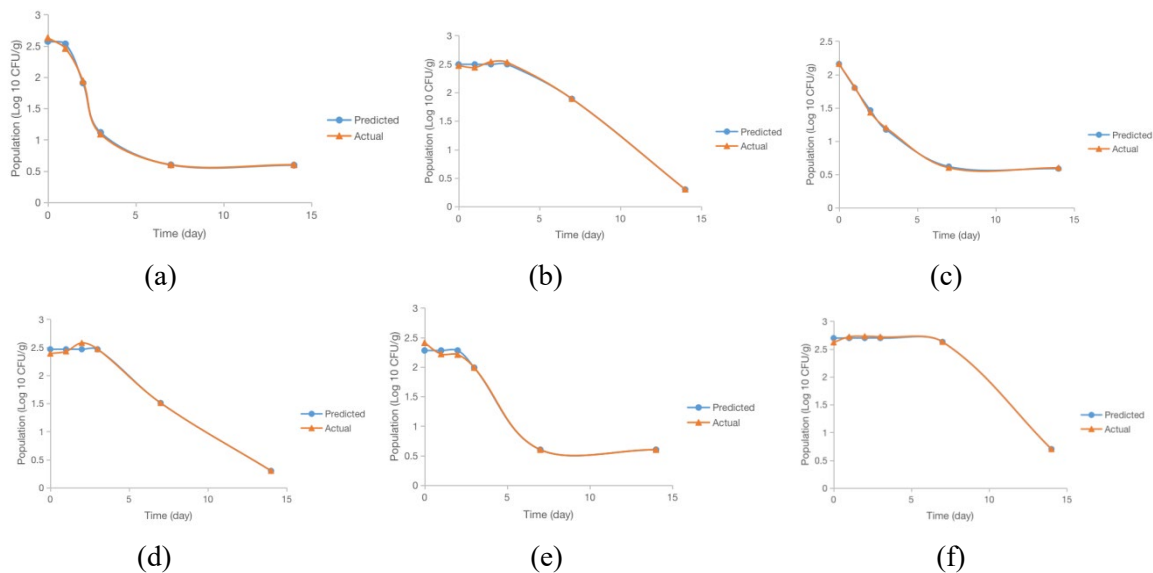


Figure 2. The predicted survival curves by modified Gompertz model and actual surviving curves of *L.monocytogenes* in butter at 21 °C with different pH: (a) pH 4.05; (b) pH 4.51; (c) pH4.58; (d) pH5.34; (e) pH5.38; (f) pH6.40.

Prediction of survival model with temperature, salt and water activity

The results of the survival model with temperature, salt and water activity are associated with the optimum survival condition to *L.monocytogenes*. And the results are similar to the result of pH, as the condition is close to the optimum condition, the inactivation rate will reduce.

Discussion

The survival of *L.monocytogenes* in butter is influenced by temperature, pH, salt content and water activity, however, still limited by many other factors. Different contaminating routes will lead to different contamination locations (Adamczewski *et al* 2022), which has a certain impact on the survival of listeria. Listeria may survive better inside butter than on the surface because of the anaerobic characteristic. But the size of moisture balls inside the butter, which may be determined by the manufacture method and type of butter, limits the survival of listeria, because *L.monocytogenes* cannot reproduce in a range close to its own size (Michelon *et al* 2016).

In this study, the survival model of *Listeria* in butter was established by taking temperature, pH, salt concentration and water activity as variables. However, the microstructures such as contamination location, initial level and droplet size were not analysed, which may have implications for the practical application of the model. These factors can be further analysed in the future.

Conclusions

This study established the surviving models of *Listeria monocytogenes* with pH, temperature, salt and water activity. As compared the accuracies of the modified Gompertz model and Baranyi model, the modified Gompertz model has higher accuracy for the primary model. The secondary models showed the relationship of parameters with inactivation rate. As the condition is close to the optimum condition, the inactivation rate will reduce. Further study should focus on oxygen condition, microstructure and other parameters.

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Pramanik Rajappa

Project Title: Co-cultivating algae and fungi for food production

Project Leader: Dr. Ronald Halim

Abstract

This study investigates the co-cultivation of microalgae and fungi. The cultures are grown in standard media, tracking the optical density, pH measurements, and cell counts. In addition, chemical analysis was performed to determine the levels of nutrients (such as nitrates, phosphates and ammonium) in the medium. The results indicated that the addition of fungi can improve microalgae growth and performance. The development of algae/fungi mixed culture can produce biomass enriched with high value metabolites and generate interesting applications in the pharmaceutical and food-production industries.



Priyadharshini Ravichandran

Project Title: Co-cultivation of microalgae and fungi

Project Leader: Dr Ronald Halim

Abstract

This study analyzes the co-cultivation of microalgae and fungi with a focus on cell wall structure. The optical density, pH measurements, and counting of the number of microalgae cells were employed to track the growth of the microalgae. Chemical analysis was performed to determine the levels of ammonium, nitrate, and phosphate in microalgae providing valuable insights into their nutrient requirements, growth dynamics, metabolic processes, and suitability for various applications. The results predicted that the co-cultivation of microalgae and fungi had a good influence on microalgae development and could be helpful for microalgae harvesting. The results of this study will add to our understanding of co-cultivating microalgae and fungi. The creation of fresh and inventive uses for microalgae may result from further study in this field, possibly advancing sustainable and environmentally friendly practices across a range of businesses.



PREDICTING THE SOLUBLE SOLID CONTENT AND MOISTURE CONTENT IN APPLES BY VACUUM DRYING TECHNIQUES USING TERAHERTZ TIME DOMAIN SPECTROSCOPY

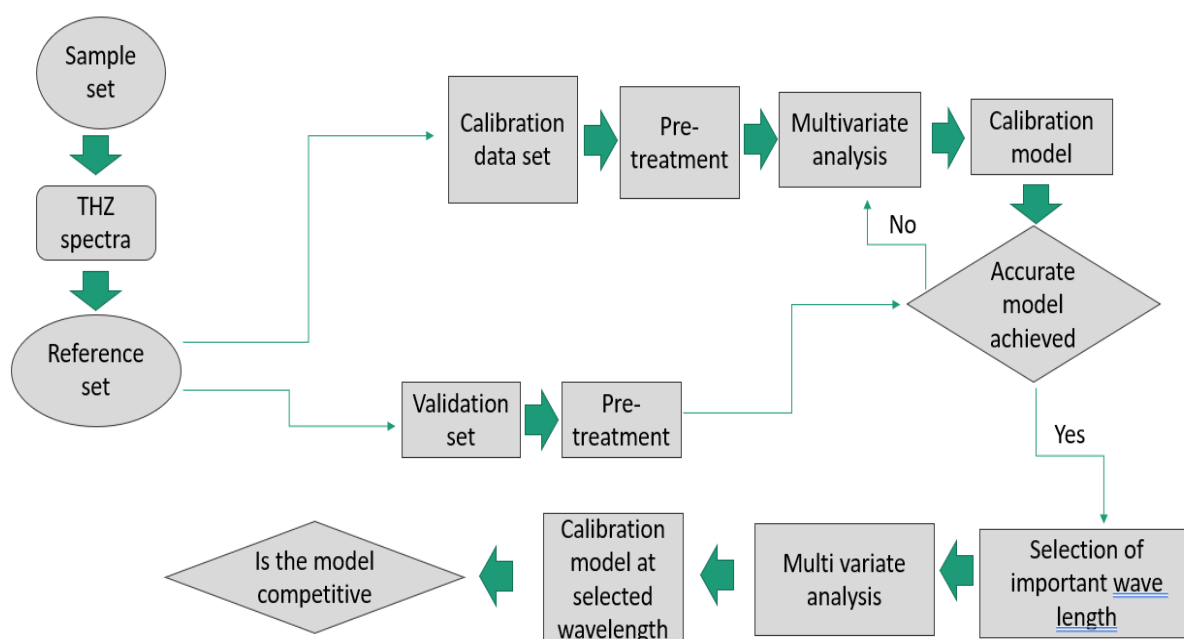
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UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

Apples are one of the most consumed products in the world. The two most important parameter that measure the shelf life and quality of the apple is moisture content and soluble solid content. This project aims to create a model that can predict the moisture content and soluble solid content using terahertz time domain spectroscopy. Several spectral pre-processing techniques such as averaging, standard normal variate, Fourier transform, principal component analysis as well as wave transform are employed to reduce signal to noise ratio and improve the quality of the data. Several algorithms are employed such as partial least square (PLS) regression which is used to make the most accurate model. The accuracy of the model is then tested using cross validation (RMSECV), root mean square error for prediction (RMSEP), standard error of prediction (SEP) and correlation coefficient.

Graphical abstract



Introduction

Apples are the most consumed fruits in the world. Apples are very nutritious and are found in every day diet and is the most widely consumed in the world. Apples are widely used in food industries to make food products and beverages. The market value of apples entirely depend on external and internal parameters. Assurance of quality of apples is integral. The two main parameters which affect the quality of apples is moisture content and soluble solid content. This where terahertz time domain spectroscopy can be used to measure these parameters (Gowen *et al* 2012).

Terahertz time domain spectroscopy is an electromagnetic spectrum between the microwaves and infrared rays. The frequency range is between 0.1 and 10 THz. It was once known as the terahertz gap and was largely ignored in mid-1990s. the low efficiency of sources and detectors of THz energy which made is a complex task to build an efficient instrumentation in this wavelength range.

The development of ultrafast lasers in the 1990 was a major turning point which lead to the modern day terahertz spectroscopy. The advantages of terahertz time domain spectroscopy is material identification, molecular dynamics, non-destructive, high sensitivity, Complementary to other techniques, high resolution and surface and interface analysis (Patocka *et al* 2020).

The objective of this study is to predict the moisture content and soluble solid content of vacuum dried apples using terahertz time domain spectroscopy.

Materials and methods

Sample preparation and drying procedure

Apples (*Malus domestica*) obtained in a local supermarket are kept in the refrigerator at 4 ± 3 °C to prevent decontamination. They were then taken out 12hours prior to the experiment to bring it to room temperature. They are washed and sliced into pieces of 5 mm thickness by using a stainless-steel slicer. The drying behaviour of 4 different shapes of apple slices were investigated in the study. Briefly, the microwave power and vacuum pressure was set at 250 W and about 20 mbar, respectively. After that, samples were taken out for Terahertz spectroscopy. Seven samples for each of the geometric shape were used in each drying batch for a certain heating time. The moisture content and soluble solid content for each whole sample were found out using oven drying method and refractometer (Pu and Sun 2016).

Terahertz time domain spectroscopy

The Terahertz (THz) spectroscopy is done using photoconductive antennas (PCAs). It is a technique for generating and detecting THz radiation. PCAs are a type of THz emitter that can produce intense, broadband THz pulses. A femtosecond laser is used to generate short pulses of near infrared range. It then passes through a beam splitter and is split into the pump beam and probe beam. The pump beam passes through the sample after passing through the emitter which produces terahertz radiation. The probe beam passes through a delay line before coming in contact with the terahertz pulse. This is to delay the probe beam before coming in contact with the pump beam in order to achieve time domain measurements. The terahertz pulse after passing through the sample will coincide with the probe beam at the detector. The pump and probe beam must coincide at the same time to produce the signal. This is because the time delay between the two beam is used to study the molecular dynamics of the sample. The terahertz spectrum is measured by delaying and measuring the signal and corresponding to each delay the entire waveform is measured. The resultant signal is a measure of both phase and amplitude of terahertz field (Gowen *et al* 2012).

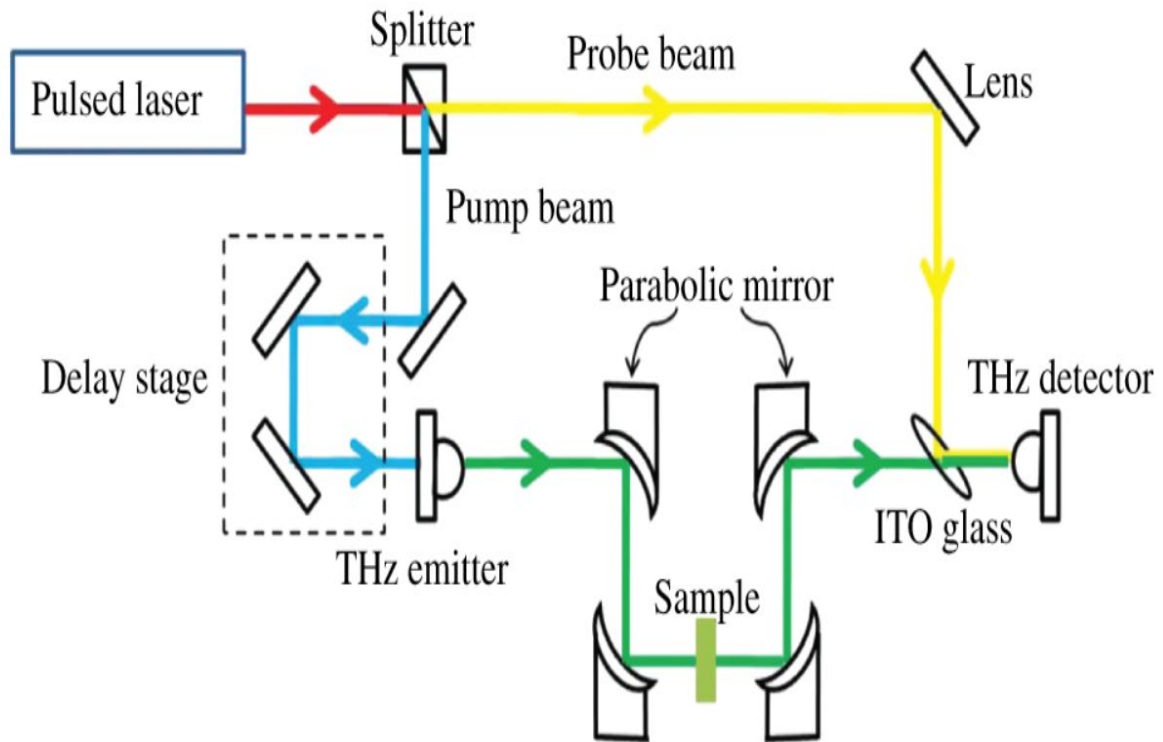


Figure 1. Terahertz time domain spectroscopy system (Yang *et al* 2018)

Data analysis and model development

After the measurements are obtained. The reference set is divided into the calibrated and validated data set. The moisture content and soluble solid content for the validation set is measured using oven drying method and refractometer. In the case of calibration data set and validation data set a series of processes must be done only after which the data can be verified. If it is successful it moves to the next phase or it is recalibrated.

Pre-treatments are a set of techniques used to process and prepare spectroscopic data before analysis. The goal of pre-treatment is to improve the quality and usefulness of the data by removing unwanted variation, such as baseline shifts and noise. One important pre-processing technique is The Fourier transform, it is a mathematical technique used to analyse and represent signals. The Fourier transform takes a time-domain signal as input and produces a complex-valued function in the frequency domain as output. The magnitude of the output function represents the amplitude of each frequency component, while the phase represents the relative timing between the frequency components. Multivariate analysis, on the other hand, is a set of statistical techniques used to analyse the relationship between multiple variables or spectra. In spectroscopy, multivariate analysis is often used to identify patterns and relationships between different spectral features, and to classify or predict the properties of samples based on their spectral data. After the model is verified with the validation data set, if successful moves to the next phase or it is re calibrated. After the verification and the accurate model has been achieved an important wavelength is selected and multi variate analysis is done on that and finally a calibration model is then prepared which is then checked for competitiveness.

Expected result

This experiment expects to see the prediction of moisture content and soluble solid content by the developed model. This experiment also proves the feasibility and effectiveness of terahertz time domain spectroscopy when measuring organic materials such as apples.

Conclusion

Predicted the moisture content and soluble solid content of vacuum dried apples using terahertz time domain spectroscopy. Measured the accuracy, completeness and robustness of the developed model. This study proves that terahertz time domain spectroscopy is a developing technology that can be used to measure the chemical characteristics and molecular dynamics of organic material. Since the radiation is of lower frequency it is far more effective than near infrared radiation in measuring organic materials. The terahertz radiation has higher absorbance to water and can be used effectively to measure the moisture content in food materials. Moisture content is one of the important parameter that determines shelf life and food quality. Thus this technology can be extensively used in the food processing industries to effectively determine the food quality of organic materials.

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CONTROLLING SPORES IN DAIRY POWDERS: SPORE POPULATION DURING PROCESSING STAGES OF SKIM MILK POWDER

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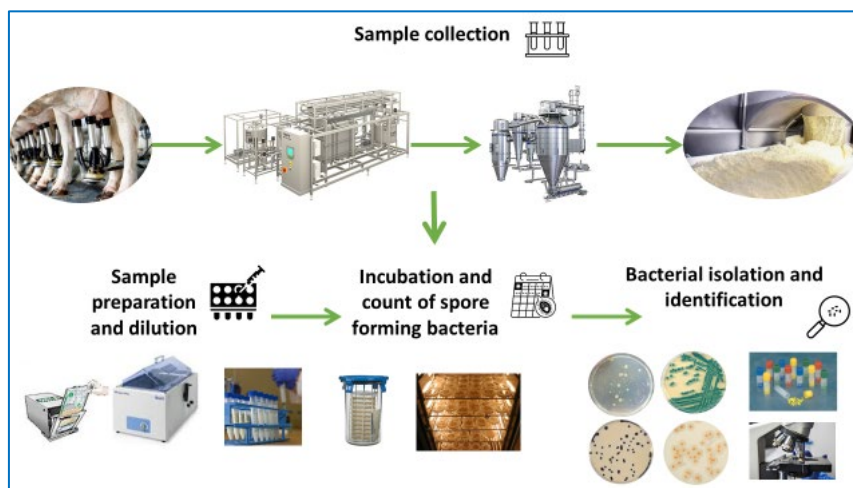
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Abstract

Spore forming bacteria cause both quality and safety issues. The *Bacillus* spores shows a dominant occurrence in dairy powders and are responsible for spoilage, decreased shelf-life and can be related to foodborne illness. *Bacillus cereus* group are the main aerobic toxin producers, Sulphur reducing spore forming bacteria are the main anaerobic toxin producers. In this project spore forming bacteria in the dairy powder process will be assessed, and the relationship between total bacteria load (total bacterial count), thermophilic, thermophilic, *Bacillus cereus* and bacterial spores will be examined. Assessment of the risk posed by spores will also be compared with dairy processing variances where data is available. Such data will contribute to knowledge gaps for dairy processors to assess and control quality and safety standards in relation to spore forming bacteria. The aim of this project will also be to assess the current quality control practices to improve microbial safety and quality issues relating to bacterial spores in dairy powders.



Introduction

Spore formers are concerning for quality and safety reasons. Because of their ability to live in harsh environments, they can develop in less competitive substrate as soon as the circumstances allow (Stenfors Arnesen *et al* 2008). Spore forming bacteria contaminate dairy products anywhere along the food chain, where the primary control points to reduce contamination are good agriculture practices applied to farm cleanliness, raw milk quality and the hygiene of processing facilities. The primary issues at the production stage involve the milking practices, bedding and bedding materials, housing system, and nutrition. The quantity of spore-forming bacteria in milk increases when silage is used as feed, but this largely relies on how the silage is stoked and whether it is carefully chosen before feeding (Borreani *et al* 2019).

The milking procedures and tools are another critical step in reducing contamination by improving the hygiene of the animal housing, and milking parlour (Borreani *et al* 2019). There are significant variations between the various bedding materials; in general, organic materials encourage the development of spore-formers (Murphy *et al* 2019). Furthermore, storage tanks, transit, and processing facilities must be clean in order to prevent contamination and biofilm development, which can contribute to recontamination (Bava *et al* 2017). The major concern related to spore formers is their resistance to a great variety of environmental conditions such as extreme high or low temperature, low pH, low water activity, radiations, desiccation, disinfectant, and chemical conservation (Jessberger *et al* 2020; Logan and Vos, 2015). The main spore formers genera found in milk and milk products, are bacillus and Clostridium (Lopez-Brea *et al* 2017).

Bacillus spp. are gram-positive bacteria, endospore formers saprophyte, facultative anaerobic (Jessberger *et al* 2020; Rahnama *et al* 2022). They are rod-shaped bacteria that can be found as individual cells or in lengthy chains. *Bacillus spp.* are the main spore forming bacteria that are related to dairy products (Gopal *et al* 2015). Within this group of bacteria is the *bacillus cereus* group of bacteria that can cause foodborne illnesses but don't need hospitalization and therefore are known to be underreported (FDA, 2005). The scientific name *Bacillus cereus sensu lato* refers to the species which is a collection of species that includes *B. cereus sensu stricto* (human pathogen), *B. thuringensis*, *B. anthracis* (human pathogen), *B. mycoides*, *B. pseudomycoides*, *B. weihenstephanensis*, *B. toyonensis*, and *B. cytotoxicus* (Cayemitte *et al* 2022). *Bacillus cereus* optimal growth temperature is between 28-35°C (FDA, 2005). According to Meng *et al* (2022) *Bacillus cereus sensu lato* strains are capable of performing proteolytic activity and this group also causes dairy spoilage. The enzymes produced are heat stable and are capable to survive at the high temperature of the heat treatments. One of the most concerning characteristics is that most of the strains in this group have psychrotrophic characteristics, and microorganisms are able to grow at very low temperature therefore can cause problems with liquid dairy products that require refrigeration (Porcellato *et al* 2021).

Bacillus cereus strains secrete a variety of extracellular enzymes that are important factors for pathogenicity, emetic (cereulide) and diarrheal (CytK, Hbl, Nhe) enterotoxins which are toxins that are linked to gastroenteritis (Vilas-Bôas *et al* 2007). The onset of the symptoms of the emetic disease caused by *B. cereus* is within 15 min to 6 h after the consumption of foods containing cereulide. The emetic syndrome symptoms are similar to *Staphylococcus aureus* poisoning and there are similarities in incubation period. 10^5 - 10^8 CFU per gram of food is enough to produce enough cereulide (5–10 µg/kg of body weight) to cause the emetic sickness. Diarrheal food poisoning can occur 8 to 16 h after consuming foods containing 10^4 – 10^9 viable *B. cereus* cells. Subsequently, the pathogen will produce and secrete enterotoxins directly in the small intestine of the consumer, which will cause a toxin-infection manifested by abdominal pain/cramps and diarrhoea. Among the diarrhoea toxigenic CytK is subdivided in two groups the CytK1 and CytK2. CytK1 is the most dangerous but rare, meanwhile CytK2 is more common but presents mild symptoms (Fagerlund *et al* 2004). The reported number of foodborne cases and outbreaks caused by *B. cereus* is frequently underestimated (underdiagnosed); not only can emetic and diarrheal syndromes be confused with those of *S. aureus* and *C. perfringens*, respectively, but also these syndromes generally clear up on their own within about 24h without requiring medical attention if there are no complications (Cayemitte *et al* 2022).

The aim of the study was to investigate the spore forming bacteria populations in dairy powders during processing and the occurrence of foodborne illness related microorganism.

Materials and Methods

Experimental plan

For 12 months, every month, skim milk powders, two associated raw materials, and two associated processed samples will be collected aseptically. Liquid samples will be tested within 24 hours, and powders samples tested within seven days. All samples will be tested in duplicate.

Bacterial methods

Total bacterial count (ISO 4833-1 2013), thermophilic bacterial count (Wehr and Frank, 2004), thermotolerant bacterial count (Wehr and Frank, 2004), and presumptive *Bacillus cereus* count (Tallent *et al* 2023) will be performed on all samples in duplicate for appropriate dilutions in the counts ranging from 10 to 300 cfu/ml.

Spore forming bacteria methods

For Mesophilic aerobic bacterial spores (MS), samples heat shocked treated at 80°C for 10 minutes, diluted if needed and 1ml pour plated using Plate Count skimmed milk agar (Merck NY USA) and incubated under aerobic conditions at 30°C for 3 days (Wehr and Frank, 2004). For high heat-resistant thermotolerant aerobic bacterial spores (TS), samples heat shocked treated at 100°C for 30 minutes, diluted if needed and 1ml pour plated using Plate Count skimmed milk agar (Merck NY USA) and incubated under aerobic conditions at 55°C for two days (Wehr and Frank, 2004). For sulphur-reducing spore bacterial count, the ISO method 15213-1:2023 to be used.

Expected results

The contamination of *Bacillus cereus* in dairy product is quite common, but low. In infant formula and milk powders the results are quite similar in all referenced papers reviewed to date (data not shown). All the results were in a range between 1 to 3 log₁₀ CFU/g. *B. cereus* spore levels in raw milk were significantly higher in summer or autumn than in other seasons in Poland (Bartoszewicz *et al* 2008), Japan (Ohkubo *et al* 2019), and Denmark (Larsen and Jørgensen, 1997). We expect similar results and predict the number of pathogenic spore-forming bacteria to be low, as in a previous study in Ireland (Li *et al.*, 2019).

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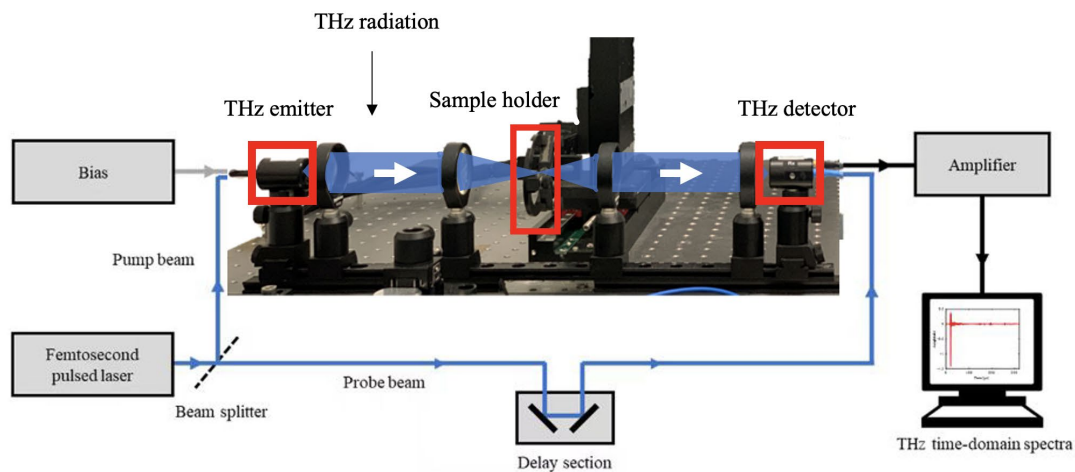
Qingxia Li, BE, MSc., PhD

Project Title: Predicting Wheat Gluten Concentrations in Potato Starch Using GPR and SVM Models Built by Terahertz Time-Domain Spectroscopy

Project Leader: Prof. Da-Wen Sun

Abstract

The purpose of this study was to explore the feasibility of terahertz (THz) spectral imaging for the detection of gluten contents in food samples. Based on the obtained 80 THz spectrum data, Gaussian process regression (GPR) and support vector machine (SVM) models were established to predict wheat gluten concentrations in 40 potato starch mixture samples. The prediction performances of GPR and SVM obtained were $R^2=0.859$ and $RMSE=0.070$, and $R^2=0.715$ and $RMSE=0.101$ in the gluten concentration range of 1.3%-100%, respectively, showing that the linear SVM algorithm had better prediction performance. The results indicated that THz spectral imaging combined with GPR could be used to predict the gluten content in food samples. It is thus hoped that this research should provide a novel technique for gluten content detection to ensure gluten-free food samples.



Selected Recent Publications

Li, Q., Lei, T., & Sun, D. W. (2023). Analysis and detection using novel terahertz spectroscopy technique in dietary carbohydrate-related research: Principles and application advance Critical Reviews in Food Science and Nutrition. <https://doi.org/10.1080/10408398.2023.2165032>.

ACCESS THE EFFECTS OF FERMENTATION TIME AND POINT OF GRASS SILAGE BALE ON GRASS QUALITY

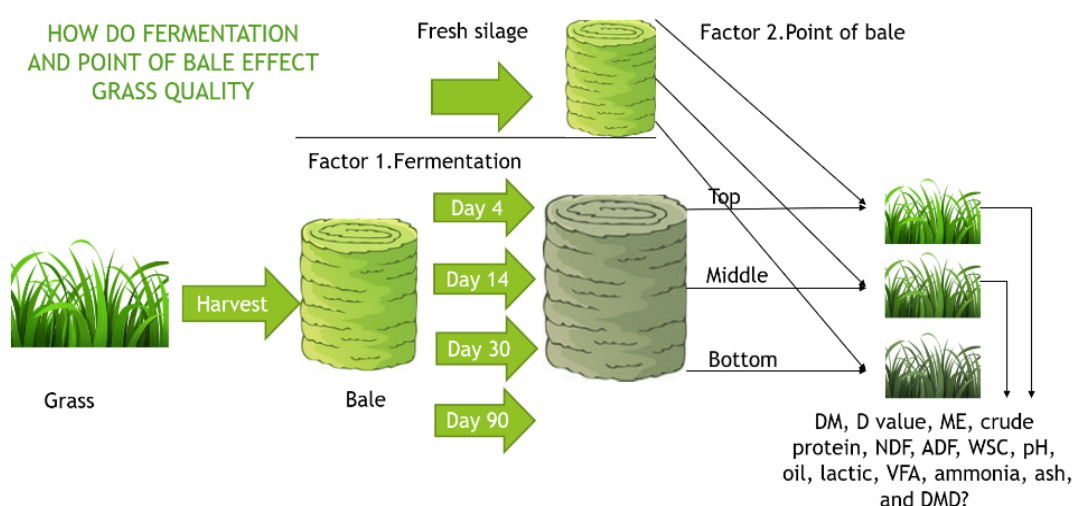
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Abstract

The core technology implementation of the Farm4more project is a green biorefinery pilot plant, with the initial stage being the establishment of an Irish grass silage feedstock supply chain. The main purpose of this study is to use NIR tools to analyze various nutritional parameters of grass silage bales, in order to investigate the effects of fermentation time and position of bale on various nutrients in grass silage bales. The experimental material is 12 packages of sustainable grass silage feed prepared from the fields of Lyons Farm in Dublin, Ireland. The 12 packages were fermented for different periods of time in a shed, resulting in four different fermentation times (0, 4, 14, 30, and 90 days) and a total of 180 samples taken from the top, middle, and bottom positions of each bale. The second experiment involves the preparation of press cake and grass juice using a hydraulic press. The third experiment is the preliminary preparation work for wet chemical analysis.

Graphical abstract



Introduction

Green grass silage is a promising feedstock for biorefineries, as it contains a range of valuable components that could be converted into a type of animal feed made by storing freshly cut grass in airtight conditions, allowing it to ferment and preserve. Near Infrared Spectroscopy (NIR) analysis can be used for feed analysis, providing a rapid and low-cost analysis of its nutritional components, and can immediately obtain results, providing a large amount of sample data for rapid analysis of forage (Shenk *et al* 1994). This can furnish pivotal information for silage quality. This thesis aims to investigate the feasibility of green grass silage biorefinery as a sustainable and will focus on the optimization of conversion processes.

The object of this study was to assess the impact of storage and point of sampling of the bale on grass quality using NIR tools calibrated against wet chemistry protocols.

Materials and Methods

Sustainable Grass silage Production

The Newcastle company, located near the UCD Lyon farm, is providing a sustainable supply chain of grass silage feed for the green pilot-scale biorefinery project. The 5.6-hectare land was reseeded three years ago with a mixture of Abergain (T), Aberchoice, Drumbo, and Buddy White Clover. This land has previously been grazed by livestock and used for the production of grass silage feed.

A total of 108 bales of grass silage were harvested on 5.5 hectares of field. 12 bales were selected at random for the bale assessment trial. Each bale has six layers of plastic packaging. Finally, these bales were transported to a warehouse in Lyons farm for storage and ensiling (Sampling period: 0, 4, 14, 30 and 90 days separate). The samples after ensiling were stored in freezer prior to analysis.

Table 1. Sward assessment for botanical composition

Species	Quadrant 1	Quadrant 2	Quadrant 3	Quadrant 4	Average	Species
Perennial ryegrass	80%	85%	90%	50%	76%	Perennial ryegrass
White clover	2%	45%	35%	45%	32%	White clover
Red clover	2%	n/a	n/a	n/a	2%	Red clover
Yorkshire Fog	n/a	5%	n/a	2%	4%	Yorkshire Fog
Creeping buttercup	1%	n/a	n/a	1%	1%	Creeping buttercup
Chickweed	n/a	2%	n/a	n/a	2%	Chickweed
Bare ground	15%	10%	10%	25%	15%	Bare ground

Near-Infrared Spectroscopy system

NIR4 Farm is a portable NIR device designed to analyse forage in real-time. With calibrations provided by Aunir. NIR4 Farm works for fresh grass and moist feeds.

Pressing Day0 and Day 30 experiment

In this experiment, silage with fermentation time of 0 and 30 days in Bale 7 was selected, and grass cake and grass juice were separated and further analyzed.

Laboratory wet chemistry analysis

By selecting 10% (18) of the total sample number for wet chemical analysis, the accuracy of the data obtained by NIRs analyzer is compared.

Vacuum sealing impact prior to freezing

In the pressing experiment, the obtained straw cake will be packaged by vacuum packaging. The purpose of this experiment is to test whether and how much vacuum packaging affects the components of silage grass.

Data analysis

The NIR4 farm was used to scan a total of 180 samples, and obtain data for 14 nutritional parameters including DM, D value, ME, crude protein, NDF, ADF, WSC, pH, oil, lactic, VFA, ammonia, ash, and DMD. The obtained data were fixed based on the latest calibration data provided by Aunir, and then imported into Excel for further analysis.

Results and Discussion

Analysis of variance

The purpose of analysis of variance (ANOVA) is to determine the extent to which two or more influencing factors affect a parameter. For the 180 original data samples of nutritional parameters taken under different conditions of time and point of grass silage bale, the ANOVA results are shown in Table 2. The significance levels are designated as ns (not significant), $P > 0.05$; *, $P \leq 0.05$; **, $P \leq 0.01$; ***, $P \leq 0.001$. Therefore, it can be concluded that the most significant influencing factor is the point of grass silage bale for parameters such as D value, ME, Crude protein, NDF, ADF, WSC, lactic acid, VFA, Ammonia, ash, and DMD.

Table 2. ANOVA table for raw data

Parameter	Source of Variation	SS	df	MS	F	P-value	F crit	significance level
DM	Time	0.744	2	0.372	0.1262	0.8815	3.0508	ns
	position	45.625	4	11.406	3.8700	0.0049	2.4264	**
D value	Time	20.956	2	10.478	2.4617	0.0884	3.0508	ns
	position	339.368	4	84.842	19.9327	0.0000	2.4264	***
ME	Time	0.086	2	0.043	0.3836	0.6820	3.0508	ns
	position	2.967	4	0.742	6.5796	0.0001	2.4264	***
Crude protein	Time	4.863	2	2.432	2.7254	0.0685	3.0508	ns
	position	166.223	4	41.556	46.5759	0.0000	2.4264	***
NDF	Time	1.272	2	0.636	0.1884	0.8285	3.0508	ns
	position	2551.655	4	637.914	188.8886	0.0000	2.4264	***
ADF	Time	4.852	2	2.426	3.7520	0.0255	3.0508	*
	position	91.850	4	22.963	35.5132	0.0000	2.4264	***
WSC	Time	1.259	2	0.629	1.7340	0.1798	3.0508	ns
	position	1923.881	4	480.970	1325.0258	0.0000	2.4264	***
Oil	Time	1.478	2	0.739	1.7959	0.1692	3.0508	ns
	position	6.811	4	1.703	4.1382	0.0032	2.4264	**
pH	Time	0.115	2	0.058	3.0571	0.0504	3.0648	ns
	position	0.042	3	0.014	0.7429	0.5283	2.6732	ns
Lactic	Time	175.015	2	87.507	1.4881	0.2296	3.0648	ns
	position	2659.009	3	886.336	15.0726	0.0000	2.6732	***
VFA	Time	29.749	2	14.874	3.5973	0.0301	3.0648	*
	position	299.772	3	99.924	24.1660	0.0000	2.6732	***
Ammonia	Time	0.183	2	0.092	1.4314	0.2427	3.0648	ns
	position	2.289	3	0.763	11.9321	0.0000	2.6732	***
Ash	Time	1.203	2	0.601	4.0141	0.0199	3.0508	*
	position	158.121	4	39.530	263.9082	0.0000	2.4264	***
DMD	Time	36.068	2	18.034	3.0780	0.0487	3.0508	*
	position	162.091	4	40.523	6.9163	0.0000	2.4264	***

Bar chart

According to the data average, standard deviation of average and four fermentation periods, draw a summary, two figures below with DM varying with sampling point and ADF varying with fermentation time as examples (Figure 1).

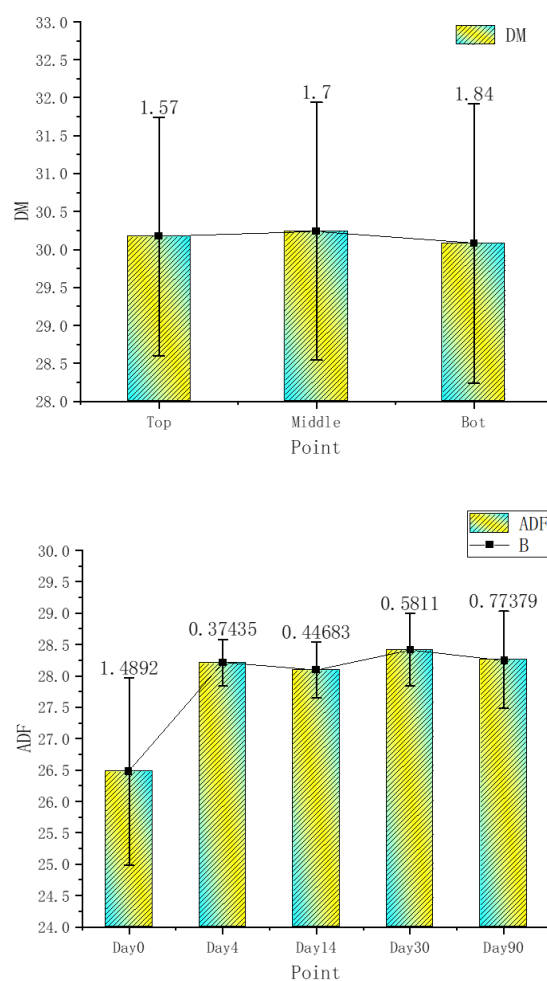


Figure 1. Average of DM and ADF value

Conclusion

The parameter data of 180 groups of forage samples at four fermentation times and three sampling positions were successfully measured by using NIR4 farm tools, and the analysis of variance was used for significant analysis. The results show that the point of silage bale is the most significant influence factor on the parameters of grass silage, such as D value, Me, Crude Protein, NDF, ADF, WSC, Lactic Acid, VFA, Ammonia, Ash and DMD. However, fermentation time only has the least significant influence on the four parameters of silage, ADF, VFA, Ash and DMD.

Reference

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HADDOCK PROTEIN RECOVERY BY ULTRASOUND-ASSISTED ISOELECTRIC SOLUBILISATION AND ALCALASE HYDROLYSIS

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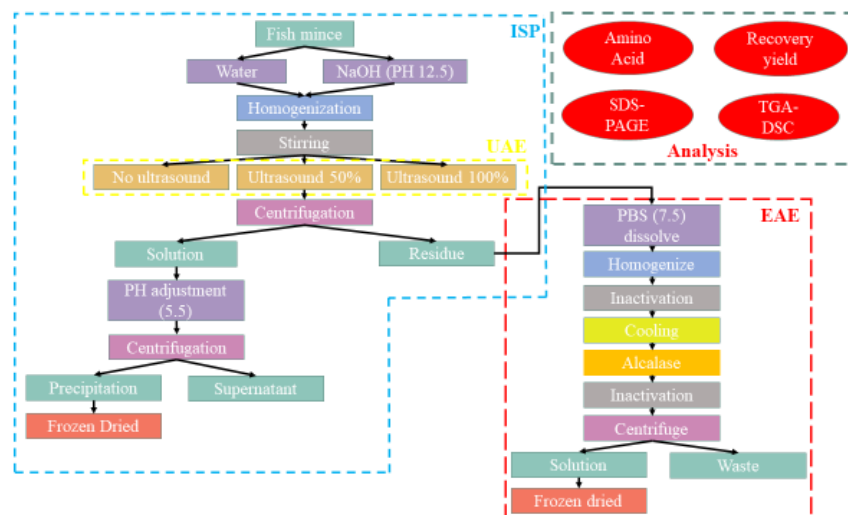
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Abstract

The valorization of fish side streams as feedstock is a burgeoning field of both academic and commercial interest on a global scale. Notably, possessing a high protein content on a dry basis, Haddock have garnered significant market demand in various industries. This study investigates the protein extraction processes by novel technology from Haddock. Use isoelectric solubilisation precipitation (ISP) under water and alkali assisted by different amplitude ultrasound (UAE), then alcalase hydrolysis (EAE) is applied to enhance the recovery. The characterization incorporates the molecular weight (SDS-PAGE), amino acid analysis, thermal stability (TGA-DSC). The research shows that 100% amplitude ultrasound-assisted alkali extraction resulted in a maximum protein recovery yield of 43.17% compared to low amplitude ultrasound treatment and water extraction in pH-shift extraction, and also has a promotion to enzymatic hydrolysis. Meanwhile, It highlights the potential of the ISP-UAE-EAE method as an innovation that residue from processing of fish can be used for isolation of high quality protein and provides a promising basis for a multiple-product fish bio-refinery, as well as for improvement with efficiency and environmental sustainability technology.

Graphical Abstract



Introduction

Under the global context of ecosystem coordination and sustainable development, the promotion of effective exploitation and valorization of natural resource is launched to release the issue of large quantity of contribution of aquaculture waste. It is estimated that 57% of the catching and processing 851,984 tons of fish resources ends up as waste (Zou *et al* 2021), consisting of head, bones, viscera, fin, skin, gut, even though which represents a valuable resource containing many high-value components including bioactive peptides, collagen. To date, the fish processing byproducts are typically reduced to compost and combustion or are landfilled, and 12% of fish produced that are used for non-food purposes, 18 million tons are diverted to fishmeal and fish oil with low profit (Siddiqui *et al* 2023).

To meet the challenge, it is desirable to develop a green technology of valorization to meet nutritional needs and reduce environmental stress by transformation of side streams for direct human consumption. Haddock is a promising source of abundant protein content, which accounts up to 70% in dry basis, with limited fat contained. Conventional protein recovery technology such as maceration, solvent extraction, steam or hydro-distillation, cold pressing and squeezing (Chemat *et al* 2017), have considerable scientific bottlenecks to overcome: more energy consumption, less efficiency and thermal sensitivity. Therefore, non-thermal processing technologies represented by ultrasound treatment attracted great attention. Sonication creates cavitation generating transient bubbles that can induce the combination mechanisms of fragmentation, erosion, capillarity, detexturation, and sonoporation (Chemat *et al* 2017), leading to changes of conformational and physicochemical properties. Meanwhile, ultrasound can be a medium to enhance the enzymatic process, which contributes to a high mass transfer from mechanical effects of ultrasound, which promotes a better interaction between enzyme and substrates, also relies on an increase of local temperature and pressure, as well as oscillatory fluid motion coming from cavitation effect (Su and Cavaco-Paulo 2021). This research investigates the optimization of protein extraction processes from Haddock using the UAE-ISP-EAE. Specifically, based on the employment of an innovative non-thermal fish protein recovery method, the functional properties and structure of fish protein under different solvents and ultrasonic intensities were evaluated. Secondly, the effect of ultrasonic treatment on alcalase hydrolysis is emphasized. Simultaneously, compare the changes of alcalase hydrolysis.

The objective of this study was to innovate the recovery of protein from Haddock waste processing technology route using Ultrasound assist pH shifting enzymatic extraction, and evaluate the properties of protein.

Materials and Methods

Ultrasound-assist Isoelectric Solubilisation Precipitation

According to former research with modifications (Álvarez *et al* 2018). Mince the fish waste sample by Mainca mincer PM-82 (Maquinaria Industria Carnica, Barcelona, Spain), keep in -20 °C fridge for further use and thaw the sample in -4 °C cold room overnight before treatment. 100 grams fish mince sample treated with alkali (pH 12.5) and waster dissolution as control, homogenize (IKA® T25 digital ULTRA-TURRAX®, Dublin, Ireland) for 30 s. After stirring 30 min under 600 rpm (VWR® VOS 40 digital, Germany), ultrasound probe (UIP 1000 hdT, Hielscher Ultrasound technology, Teltow, Germany) is employed with different amplitudes (50%, 100% and no ultrasound) for 10 min. Centrifuge and precipitate the protein in supernatant by shifting the pH to 5.5, and the residue was kept under -20 °C fridge for further use.

Enzymatic Hydrolysis

According to researches (Alavi *et al.*, 2019; Hauet *et al.*, 2022) with modifications. The residue is thawed under 4 °C cold room overnight. Dissolve the residue in water with ratio of 1:4 and homogenize for 30s. Treat with 90 °C water bath for 10 min deactivation to eliminate the effects of endogenous enzymes. Adjust pH to 8.0 and incubate the substrate in orbital shaker (Model SHKE6000-1CE, Thermo Scientific, U.S.A.) under 55°C and 130 rpm for 15 min. The alcalase (Bacillus licheniformis) (Merck, Darmstadt, Germany) with activity unit of 2.972 U/ml is added with 8% (v/w) of sample and continue the incubation for 2h. Placed in 90°C water bath for 10 minutes to stop the enzymatic reaction. The mixture is then centrifuged and the soluble protein layer is freeze-dried kept under 4 °C.

Table 1. Classification of the extraction samples.

Group	Solvent	Ultrasound Amplitude	Residue
1	Water	0	Alcalase
2		50%	
3		100%	

4	Alkali (pH 12.5)	0	
5		50%	
6		100%	
7	-	-	

Nitrogen Analysis

The nitrogen analysis is determined using the Dumas method (LECO FP628, 3000 Lakeview Avenue, St. Joseph MI 49085) and the protein factor is set as 6.25.

SDS-PAGE

SDS-PAGE of protein extracts are performed using the former method (Sun *et al* 2021). Each sample (10 µg) was loaded onto 8% polyacrylamide gel. After electrophoresis, the gel was stained with Coomassie brilliant blue (CBB) R-250 and destained using 30% (v/v) methanol and 10% (v/v) HAc. Quantitative analysis of protein band intensity was done using Quantity One software (Version 4.6.2, Bio-Rad, Hercules, CA, USA).

TGA-DSC

According to the research (Ricci *et al* 2018). Thermogravimetric analysis was conducted on a TA instruments TGA Q50 series. DSC analysis was conducted using TA instruments DSC Q2000 series. Samples were analyzed using a heating ramp of 10 °C/min to 550 °C with nitrogen at a flow rate of 50 mL/min as the carrier gas to obtain the decomposition temperature of the polymer.

Amino Acid Analysis

Amino acids were analyzed using an amino acid analyzer (Jeol JLC-500/V, Jeol Ltd., Garden city, Herts, UK) based on the research (Zhang *et al* 2021).

Data analysis

All experiments were replicated two times and the analyses were carried out on all samples. Results are expressed as mean ± standard deviation (SD). Difference between samples were analyzed using analysis of variance (ANOVA) performed using SPSS. Values were considered significant at $p < 0.05$.

Results and Discussion

Protein recovery yield by EAE-ISP

The research found that 100% amplitude ultrasound-assisted alkali extraction resulted in a maximum protein recovery yield of 43.17%, significantly higher than the water recovery yield of 5.73% under the same conditions. Furthermore, the study revealed that ultrasound treatment significantly enhanced the protein recovery yield (36.28% for alkali with no ultrasound), with the yield gradually increasing with the enhancement of amplitude (42.56% for alkali with 50% ultrasound).

Table 2. Protein Recovery by UAE-ISP

Groups	NaOH (pH 12.5)	Water
Ultrasound 0%	36.28 ± 0.04%	1.45% ± 0.71%
Ultrasound 50%	41.56% ± 1.94%	1.42% ± 0.30%
Ultrasound 100%	43.17% ± 0.78%	5.73% ± 1.14%

Prediction of protein properties

SDS-PAGE will show the hydrolysis of macromolecular proteins by alcalase resulting in a large number of small molecule peptide chains and amino acids, and compare the protein composition of various molecular weight under different ultrasonic treatments and solvents. Amino acid

analysis will reveal the amino acid composition of haddock in different processes respectively. TGA-DSC will obtain the changes of protein powder mass and heat absorption during the heating process. The analysis above collaboratively have decision on the protein constructive and functional properties.

Conclusions

It highlights the potential of the ISP-UAE-EAE method as an innovative technology for the extraction of protein from Haddock, which can then be applied to the recovery of residues. These findings have significant implications for the development of advanced protein extraction methods that are more efficient and environmentally sustainable, addressing critical challenges in the food industry.

Acknowledgements

The authors would like to acknowledge University College Dublin (UCD) and China Scholarship Council (CSC) for their support of this research.

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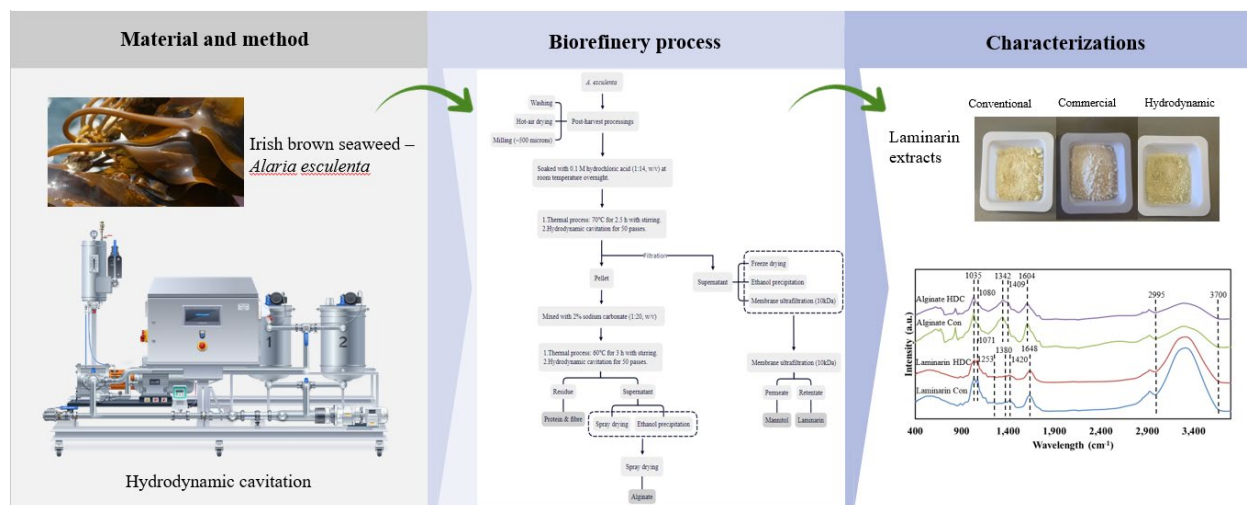
Xianglu Zhu, B.E., M.Sc., Ph.D.

Project Title: Hydrodynamic cavitation for brown seaweed in a cascading biorefinery model for laminarin, alginate and protein extraction

Project Leader: Prof. Da-Wen Sun

Abstract

The global market of seaweeds was USD 10.31 billion in 2020 and estimated to reach around USD 22.13 billion by 2024. This project investigates a novel biorefinery process designed for the extraction of valuable compounds from brown seaweed *Alaria esculenta* using hydrodynamic cavitation (HDC). A two-stage process was developed to maximise the value of seaweed biomass by control of the processing time, solvent selection and HDC conditions to extract laminarin, alginate, mannitol and protein in a cascading manner. After the first extraction stage using 0.1 M HCl, membrane ultrafiltration was employed to separate laminarin and mannitol. The purity of the laminarin and mannitol obtained was $86.57\pm3.72\%$ and $40.49\pm2.78\%$ with recovery rates of $55.55\pm3.10\%$ and $75.90\pm4.49\%$ respectively. Ethanol precipitation was then carried out to recover sodium alginate after the second extraction stage process using 2% Na_2CO_3 (w/v). The sodium alginate purity extracted by HDC-HDC was $88.98\pm4.70\%$ with a recovery rate of $65.13\pm5.14\%$. The remaining residue after the biorefinery process had an enriched protein content of $17.19\pm1.33\%$. This study demonstrates that a HDC assisted biorefinery process can significantly ($P<0.05$) reduce energy consumption. The laminarin extracts obtained were characterised by FT-IR spectroscopy and measurement of both antioxidant and anti-inflammation activities. The laminarin extracted in this study was shown to have identical bioactive activities as the commercially available samples.



Selected Recent Publications

- Zhu, X., Patange, A.D., Macori, G., Sun, D.W. and Tiwari, B.K., (2022). Impact of high pressure treatment on shelf life and microbial profile of wild harvested *Ascophyllum nodosum* and aquacultured *Alaria esculenta* during storage. *LWT*, 170, p.114022.
- Zhu, X., Healy, L.E., Sevindik, O., Sun, D.W., Selli, S., Kelebek, H. and Tiwari, B.K., 2022. Impacts of novel blanching treatments combined with commercial drying methods on the physicochemical properties of Irish brown seaweed *Alaria esculenta*. *Food chemistry*, 369, p.130949.

INVESTIGATING THE MICROPLASTICS AGING UNDER SOME CONDITIONS AND THE FATE OF MICROPLASTICS IN FOOD BIOLOGICAL SYSTEMS

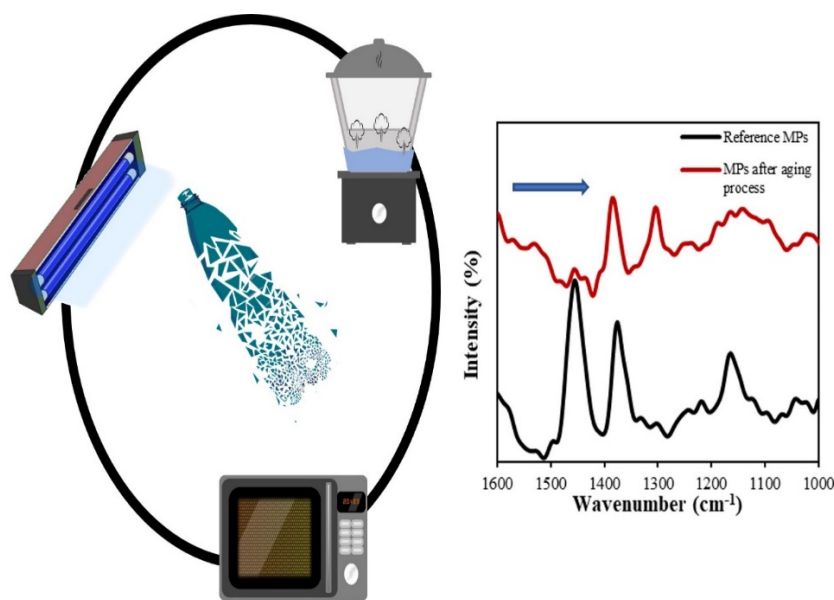
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Abstract

Microplastics (MPs) are small plastic particles less than 5 mm in size that have become ubiquitous environmental pollutants. They are now found in various environments, including water bodies, air, and even in the food chain. While their effects on organisms and ecosystems are widely studied, less is known about the aging of MPs and their potential to degrade over time. The aging process of MPs is a complex and dynamic process that is still not fully understood, and further research is needed to better understand the fate and behaviours of MPs. In the present project, microwave, UV, and steam treatments will be applied as aging processes to determine their effects on the chemical structure of MPs. Also, the study focuses on examining how food matrices can impact the aging process of microplastics, as well as the potential interaction between food components and microplastics. Preliminary data revealed that MW treatment could cause changes in the morphological and chemical structure of MPs.

Graphical Abstract



Introduction

Plastics have been considered a new type of persistent contamination as they can persist in water for hundreds or even thousands of years. Despite this situation, it is expected that plastic production will reach 500 million tons in 2005 (Ainali *et al* 2021). Plastic wastes can be degraded to microplastic particles (MPs) ranging in size between 100 μm and 5 mm in the environment (Kadac-Czapska *et al*

2023). It is known that air and water sources are contaminated with MPs, and research has also shown contamination of MPs in the food supply chain. MPs in beverages and processed foods, resulting in 39,000–52,000 MPs particles typically consumed per person annually (Vitali *et al* 2023).

Plastic materials used in food packaging have the potential to release in the form of microplastics into food and then into the environment. Under certain storage or processing conditions, the reactivity and toxicity of the aged MPs might alter. Food packaging plastics can be exposed to ultraviolet (UV) radiation in bad storage conditions or after they become plastic waste in nature. UV oxidation is highlighted as one of the main ways MPs aging in the environment. The surface of MPs would become fragile with pores and holes, and it would provide more accessible reaction sites, promoting polymer aging. These activities might cause changes in the characteristics of MPs and the production of secondary MPs, which affect their fate and environmental hazards (Wu *et al* 2021).

Microwave ovens are one of the most used kitchen appliances, and food is frequently heated in the microwave while still in the microwavable plastic polymer container. Despite these containers being considered microwavable, due to be reached high temperatures in a short time with microwaves, this process facilitates the migration of MPs. In addition, reusable food packages could cause unpredictable polymer behaviour due to degradation (Alin and Hakkarainen 2012). Steam heating is preferred for healthy food and also for sterilization of containers such as baby feeding bottles. Su *et al* (2022) show that a large number of MPs in different size ranges were formed by steam-induced degradation during the sterilization of infant feeding bottles.

In addition to UV or other heating effects, the interaction between different food components and plastic material might alter the migration and degradation profiles of MPs. Alin and Hakkarainen (2011) demonstrated that the water solubility of the migrants constricted the migration into aqueous food simulants and that the fatty food model displayed considerably improved overall migration values in comparison to the other models.

One of the objectives of this study is to determine the effects of UV treatment, microwave heating, and steam on MPs aging. In addition, the effect of food matrices on this process and the interaction of food components and MPs will be investigated.

Materials and Methods

Polypropylene (PP), Polystyrene (PS), Polyethylene (PE), Polyethylene Terephthalate (PET), and Nylon, which are the most used plastic materials for food packaging, were preferred as MPs samples in this study. Different MPs solutions of the same concentration will be prepared in DI water for further analysis.

Microwave (MW) treatment

A household MW combination oven (Panasonic NN-DS59NB, Panasonic Connect Co, Japan) will be used with different output powers between 100W and 1000W. Microwaves ranging from 0 to 15 min will be applied, and to prevent overheating or boiling of the samples, a single exposure will be taken for 30 s followed by a cooling step. After treatment, aliquots of samples will be dried on appropriate glass slide and analysed via optical photothermal infrared microspectroscopy (O-PTIR, Mirage, Photothermal Spectroscopy Corp, USA) or QCL-IR Microscopy (Spero Chemical Imaging Microscope, Daylight Solutions Inc., USA). The obtained IR spectra of MW-treated MPs samples will be compared with untreated control samples to understand the potential change in the chemical structure of MPs. In addition, MW will be applied to slides of untreated control samples and the same particle will be analysed after MW treatment. The zeta potential of MW-treated and control samples will be analysed by a zeta sizer.

UV treatment

A UV lamp will be utilized for exposure to UV radiation on MPs samples. To understand the aging effect of this process, MPs solution in glass Petri dishes will be placed under UV irradiation. The

treatment will be applied according to (Sun *et al* 2022). Control samples were covered with aluminium foil in a dark location. The chemical structure of samples will be analysed with the same method as the previous session.

Steam infusion

The steam mode of the combination oven will be used for analysis, and the effect of steam infusion will be investigated by following Su *et al* (2022). The IR spectra of steam-treated and control samples will be analysed by O-PTIR or QCL-IR Microscopy.

MPs and food model interaction

Bovine serum albumin, casein; fructose, glucose, sucrose, and maltose; olive oil, and fish oil will be used as food models. The concentration of the ingredients in food models will be equivalent to that of MPS solutions, and they will be incubated for one hour at room temperature with MPs samples. Enzymatic or chemical extraction will be used (Sridhar *et al* 2022) if considered required, and following that, the chemical structure of MPs will be analysed.

In addition, the interaction between MPs and food models will be investigated with further analysis. Morphological characterisation and corona structure will be investigated via scanning electron microscopy. The zeta potential and hydrodynamic diameter of the samples will be evaluated by a zeta sizer.

Results and Discussion

The analysis planned to be carried out with the PhD project is stated in the material method section. Preliminary data obtained as a result of MW treatment of PP MPs were presented in this section.

MW treatment on PP MPs samples

1000 $\mu\text{g/mL}$ PP MPs (mean diameter: 90 μm) solution (w/v) in DI water was prepared, and 540 W MW was applied to this solution for 10 min. IR spectra, which were obtained by QCL-IR Microscopy, of MW-treated and control samples are shown in Figure 1. Characteristic peaks for PP polymer were found at 1456 cm^{-1} and 1376 cm^{-1} related to CH_3 symmetrical bending; 1164 cm^{-1} related to C-H bending and CH_3 rocking; 996 cm^{-1} and 972 cm^{-1} attributed to C-C bond stretching. After MW treatment, some peaks shifted slightly, and the 996 cm^{-1} and 972 cm^{-1} peaks were missed.

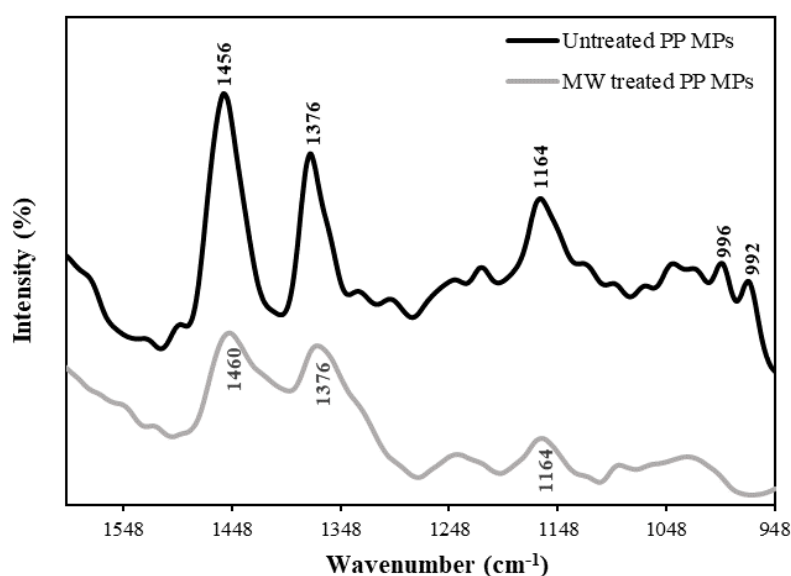


Figure 1. IR spectrum of control and MW treated PP MPs samples.

The particle area and circularity of the PP MPs sample were also investigated (Table 1). After the MW treatment, the area of the particles decreased while their circularity increased.

Table 1. Area and circularity value of samples

	Sample Number	Area (μm^2)	Circularity
MW treated PP MPs	1	222	0.92
	2	92	0.97
	3	296	0.58
	4	222	0.78
Untreated PP MPs	1	3310	0.74
	2	7026	0.76
	3	1220	0.46
	4	1646	0.48

Conclusions

The results obtained from the preliminary experiment showed that there might be a change in the characteristic properties of PP MPs after MW treatment. In order to interpret the results more accurately and to perform statistical analysis, further analysis will be progressed.

Acknowledgements

This work has been supported by Science Foundation Ireland (SFI)-Irish Research Council Pathway Programme under Proposal ID 21/PATH-S/9290.

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THE EFFECT OF UV AGEING ON SPECTRAL PROPERTIES OF POLYSTYRENE MICROPLASTICS

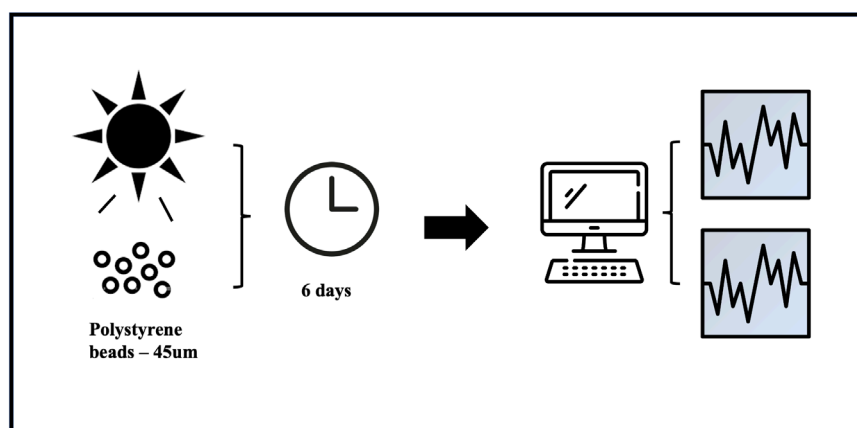
Raphaela O.G. Ferreira, Aoife Gowen, Junli Xu

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

Polystyrene is a commonly used polymer for food containers and office supplies that can end up in the environment and undergo various degradation processes, including photodegradation. This study aimed to investigate the effects of UV ageing on the spectral properties of polystyrene microplastics through spectroscopic analysis. Polystyrene beads with a diameter of 45 μm were diluted, aged for six days under UV light exposure, and analysed using mIRage® IR Microscope. The acquired spectra underwent pre-treatments such as Standard Normal Variate (SNV). Principal Component Analysis (PCA) was used to visualize similar observations and differences due to UV weathering. The results indicated a decrease in the size of the polystyrene beads, a significant difference in absorbance intensity between treated and untreated samples, and a trend of cluster separation after UV light exposure. These observations suggest that UV ageing can induce changes in the structural and spectral properties of polystyrene beads and highlight the potential environmental impact of polystyrene degradation.

Graphical Abstract



Introduction

Plastics in the environment undergo slow degradation leading to fragmentation into fibres, granules, flakes and spheres with a diameter or length of less than 5 mm, called microplastics (MPs) (Elizalde-Velázquez *et al* 2021).

Being one of six plastic types widely used in Europe, polystyrene (PS), a small organic compound with the chemical formula $\text{C}_6\text{H}_5\text{CH}=\text{CH}_2$, is found in packaging materials and office supplies because of its high transparency, durability, and easy dyeing (Plastics Europe, 2022; Rocha-Santos and Duarte, 2015).

When PS is utilized as a disposable food container for immediate use, it becomes more vulnerable to the effects of outdoor weathering. Once exposed to the environment, MPs degrade first at the polymer surface, which is exposed and available for chemical or enzymatic attack, eventually leading to embrittlement and disintegration.

Infrared spectroscopy has become one of the most powerful and versatile tools at the disposal of modern bioscience. Spectroscopy is a non-invasive technique with rapid measurement, which enables the identification and differentiation of the internal chemical composition of the detected object (Beć *et al* 2020).

The aim of the study was to investigate the effects of UV weathering of polystyrene MPs using spectra analysis.

Materials and Methods

Monodisperse polystyrene beads of 45µm diameter were purchased from Polysciences, Inc and were diluted in sterile distilled water (Gibco) for a final concentration of 200 µg/ml. 1 ml of the solution was transferred to a petri dish and was artificially aged for six days exposed in UV light at room temperature. After that, polystyrene beads were resuspended in 1ml of sterile distilled water and 20 µL of this solution was deposited on a cover slide and dried at room temperature. Control samples were also prepared following the methodology described above.

Samples were analysed in mIRage® IR Microscope, Photothermal Spectroscopy Corp. for image acquisition, beads measurement (10x and 40x), and spectra acquisition - with 3 spectra pixels of each bead. The extracted spectra (n=12) were then imported into the Matlab software (The MathWorks Inc., Natick, MA, USA) for further analysis. The spectra were subjected to a few pre-treatments including SNV (Standard Normal Variate). Principal Component Analysis and Score Plot were also calculated.

Results and Discussion

The ageing of Polystyrene MPs by the UV treatment was explored in this study. After UV exposure and further recovery of PS beads, the solution of PS MP beads showed a yellow aspect – it was transparent before UV ageing, confirming the colour change described by Beć *et al.* (2020). PS MPs beads maintained spherical after UV ageing. However, after PS MPs were measured in O-PTIR software (N=3), a difference in their average diameter was observed – 39.56µm in control samples against 37.73µm in UV treated PS beads, suggesting that the PS MPs shrunk a few micrometres after the UV exposure. According to Beć *et al.* (2020), shrinkage is a common consequence of UV degradation.

Mean Spectra

To decrease spectral variability, standard normal variate (SNV) can be applied to the data to correct spectral errors ranging from particle size to surface texture (Zhu, 2019). The mean spectra of polystyrene 45um before and after UV exposure (black and red spectra, respectively) for 6 days was reported in the wavenumber range 1800-800nm (Figure 1).

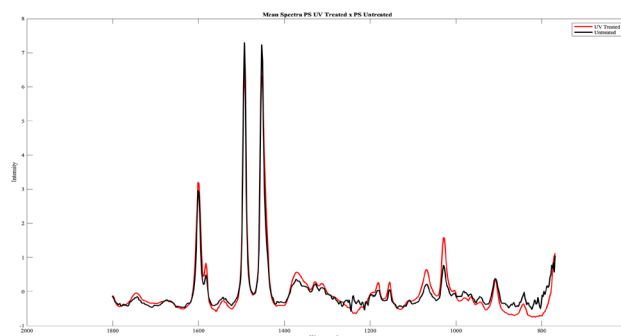


Figure 1. Mean Spectra of Polystyrene 45µm UV treated for 6 days and Polystyrene control 45µm (untreated) - after preprocessing with SNV plotted against wavelength (nm).

Important regions attributed to specific functional groups for the prediction of the Polystyrene are listed in Table 1. These regions are matching with the IR spectra previously described in Al-Kadhemy *et al* (2016) and Battulga *et al* (2022).

Table 1. IR characteristic peaks of Polystyrene 45µm before and after UV treatment

Regions and functional groups	Polystyrene 45µm Experimental peak
C–H out phase bend (625-970) nm	906, 842
C–O stretch (880-1000) nm	906
CH ₂ bending (1300-1380) nm	1311,1373
C≡C stretch (1550-1610) nm	1539,1600
C≡O (1550-1750) nm	1539,1600

UV treatment resulted in a significant reduction in intensity absorbance at 842nm, indicating chemical transformation or degradation of the stretching of C-H bonds. As discussed in Beć *et al* (2020), UV light has been shown to initiate photodegradation in certain molecules, causing alterations in their spectral characteristics, such as the breakage of chemical bonds, modification of electronic structure, or crosslinking between molecules, which can lead to a decrease in concentration or shift in the absorption spectrum of the absorbing species.

PCA Score Plot

The score plots help to visualize similar observations and differences due to UV weathering. The first two scores - PC1 (43.67%) and PC2 (24.54%), explained the greatest variation in the data and were plotted in Figure 2. The observed separable trend between the black and red clusters (untreated and treated, respectively), reinforces the differences previously identified in the spectra and optical images.

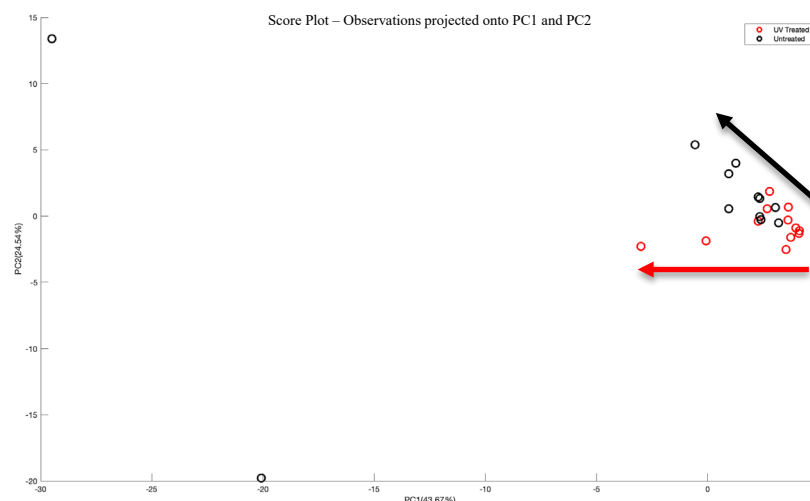


Figure 2. Principal Component scores (PC1 vs PC2) for Polystyrene 45um UV treated for 6 days and Polystyrene control 45 um (untreated) spectra after SNV.

Conclusions

This study investigated the effects of UV exposure on PS beads. The beads underwent degradation, as evidenced by a decrease in diameter. Analysis of the mean spectra showed a significant difference in absorbance intensity between the UV-aged and control samples. PCA demonstrated a trend of separation between the two sample groups, indicating significant changes to the structure and optical properties of the beads. Future experiments could extend the duration of UV exposure and test different bead sizes to confirm these trends.

Acknowledgements

This research has been undertaken with the financial support of Science Foundation Ireland (SFI) – Irish Research Council Pathway Programme – Proposal ID 21/PATH-S/9290.

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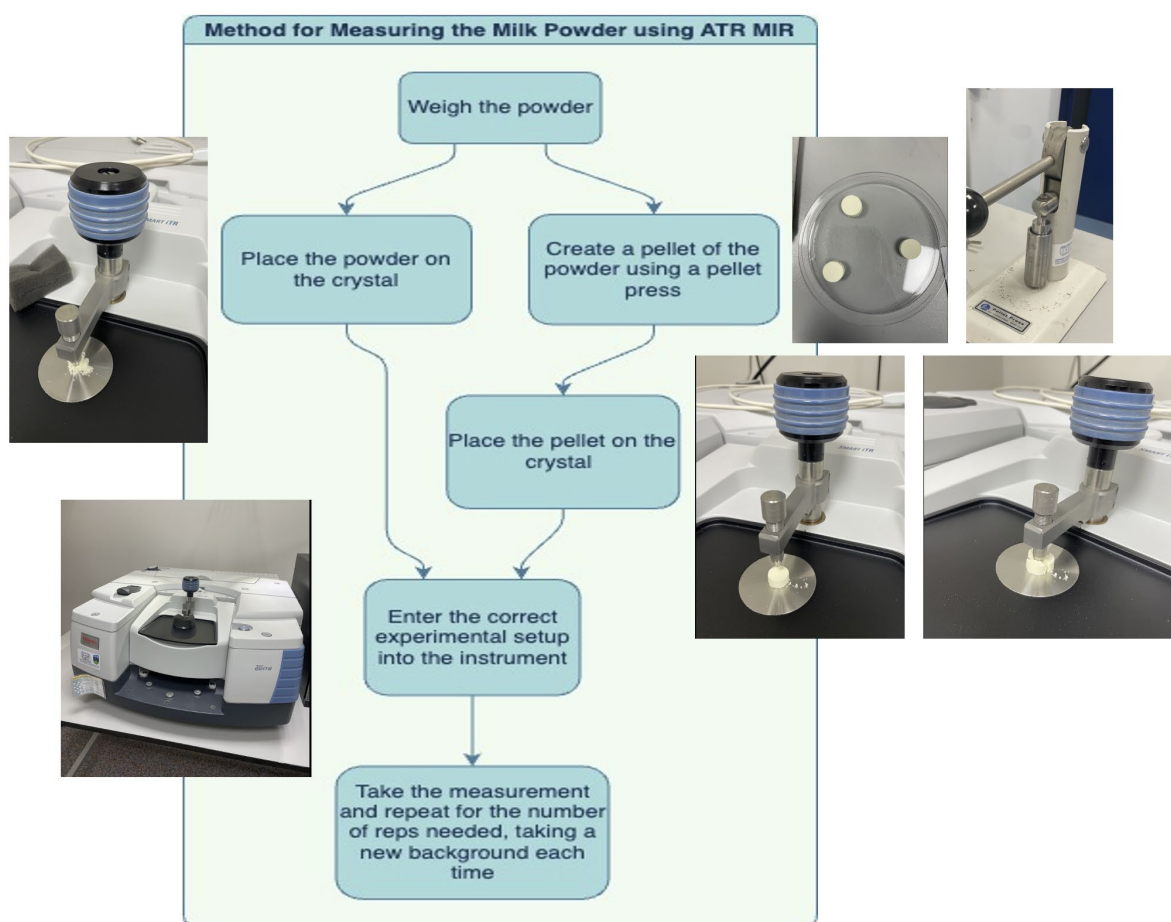
Áine Ní Fhuaráin, BSc, MSc

Project Title: An investigation into the influence of sample presentation in the measurement of milk powder using Attenuated Total Reflectance (ATR) Mid-Infrared (MIR) spectroscopy and Principal Component Analysis (PCA).

Project Leader: Professor Aoife Gowen

Abstract

The global production of Whole Milk Powder (WMP) and Skim Milk Powder (SMP) is predicted to increase between 2019 and 2028. Milk powder is used for recombination and reconstitution as well as in other food products. Spectroscopy can be used as a tool to analyse the properties of milk powders and is advantageous because it is a rapid, inexpensive, robust and non-destructive technique. Therefore, a chemometric model using spectroscopy to determine milk powder types would be beneficial to industry. A development of a reliable method with which to analyse various milk powders using ATR MIR spectroscopy is studied, which would be useful to ensure the properties of the milk powder are consistent. Samples of WMP, SMP, protein powder and infant formula powder are analysed, using two sample presentation methods: 1) pelletizing the powder before placing it on the ATR crystal and 2) measuring a mass of 0.4 g loose powder on the crystal. Multiple replicate spectra were taken of each sample using each sample presentation method with a Nicolet IS50 FTIR (Thermo Fisher Scientific). PCA was performed using MATLAB to determine which method has better repeatability. Future work will include using chemometrics to create a predictive model for milk powder types.



INVESTIGATING THE RELEASE OF MICROPLASTICS FROM BAKEWARE DURING OVEN HEATING

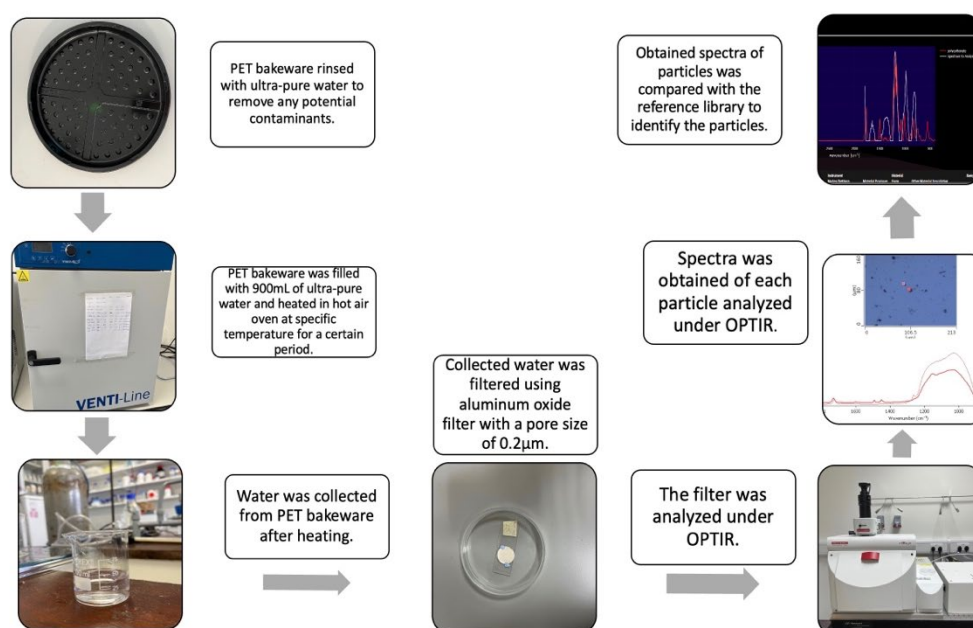
Kunjan Govil, Aoife Gowen, Junli Xu

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

The purpose of this study is to examine the effects of heat treatment on the release of microplastics and to develop a method for identifying microplastics in bakeware made of polyethylene terephthalate (PET). The work is important because it adds to our expanding understanding of the possible health dangers posed by plastics in materials used in food contact and offers information that may help shape future laws for safer food packaging. PET bakeware will be filled with 500 ml of water and rinsed with ultra-pure water as part of the experiment before being heated in a hot air oven at a specific temperature for a predetermined amount of time. After heating the PET bakeware, water will be extracted from it. Using aluminium oxide with 0.2 micron pore size as a filter, the water will be collected. After that, the filter will be subjected to optical photothermal infrared (O-PTIR) spectroscopic analysis, and the spectra that are produced will be recognized using a reference library. In addition to identifying the ideal settings for decreasing microplastic release, the project hopes to quantify the number of microplastics released under various time and temperature conditions.

Graphical Abstract



Introduction

Due to their many attractive qualities, including durability, lightweight, and affordability, plastics have become a necessary component of our daily life. Microplastics, a new type of environmental pollutant, have emerged as a result of incorrect disposal and increased plastic manufacture. Small plastic particles known as microplastics, which are less than 5 mm in size, can harm the environment and human health by getting into the food chain (Geyer *et al* 2017). According to estimates, plastic debris contributes to

around 90% of the microplastic pollution in the ocean, with primary microplastics making up the remaining 10%. (Thompson *et al* 2004).

Because of its beneficial qualities, polyethylene terephthalate (PET) is a type of plastic extensively utilized in the production of food contact products such as bottles, trays, and bakeware. However, a technology for detecting microplastics from PET bakeware needs to be developed because their presence and potential release during the preparation and storage of food could pose a serious health danger (Rist *et al* 2018).

Optical photothermal infrared (O-PTIR) spectroscopy, a cutting-edge method, has the ability to find microplastics in a range of matrices, including materials used in food contact. O-PTIR is based on the idea that when infrared radiation is absorbed by a sample, it causes an increase in temperature that is proportional to the amount of radiation absorbed. This technique has been used to find microplastics in environmental samples like wastewater and marine sediment (De Frond *et al* 2019). Nevertheless, no research has been done on using O-PTIR to detect microplastics in PET bakeware, and it is unclear how heat treatment affects the release of microplastics from PET bakeware.

The study's findings may help shape future regulations for safer food packaging and offer important new information about the potential health concerns associated to the use of plastics in food contact materials.

The goal of this study is to create an O-PTIR spectroscopy-based technique for detecting microplastics in PET bakeware. The effects of heat treatment (temperature and exposure time on the release of microplastics from PET bakeware correspond to what we are attempting to determine.

Material and Methods

Sample Acquisition

PET bakeware samples were ordered online and thoroughly inspected upon delivery to prevent damage and contamination. Due to their widespread use in food packaging and the possibility that they could release microplastics when heated, PET bakeware samples were selected as the study's main subject.

Rinsing of PET bakeware

Ultra-pure water was used to rinse the PET bakeware before usage in order to get rid of any potential impurities that might affect the analysis.

Heat treatment of PET bakeware

The PET bakeware was filled with 900mL of ultra-pure water and covered with a glass lid to prevent contamination and water loss during heating. It was heated in a hot air oven at 220°C for a certain period of time to stimulate real-world use conditions.

Table 1. Heat treatment of PET bakeware at different times.

Temperature (°C)		Time (min)	
220		20	
220		40	
220		60	
1 PET bakeware	Temperature 220°C	Time 60mins	*Result will be recorded after 3 rd heating cycle

*One piece of PET bakeware underwent a heat treatment in a hot air oven for 60 minutes at 220°C. Three times this heat treatment was performed, and following the third heating, water from the PET bakeware was collected for filtration and analysis.

Collection of water sample

After heating, the PET bakeware was allowed to cool for a few minutes before collecting the water. The water collected from the PET bakeware was carefully put into a flask, and the volume was measured. Also, any water that was spilled was noted.

Filtration

To remove microplastics from the water, aluminium oxide filters with 0.2 μm pore sizes were used to filter the collected water samples. As aluminium oxide filters have a high adsorption capacity and can efficiently retain particles of all sizes, including microplastics, they were used for filtration.

Spectroscopy and Identification

OPTIR (Optical Infrared) spectroscopy was used to examine the aluminium oxide filter. For each observed particle, spectra were produced. To ascertain whether there were any microplastics in the filter, the produced spectra were then matched to a reference library of known microplastics.

Results and Discussion

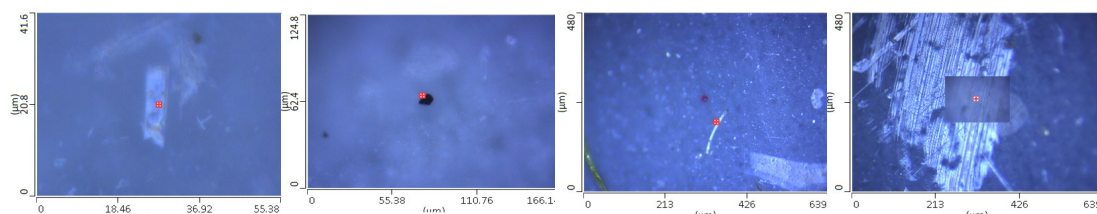


Figure 1. Particles 1,4,8, and 9.

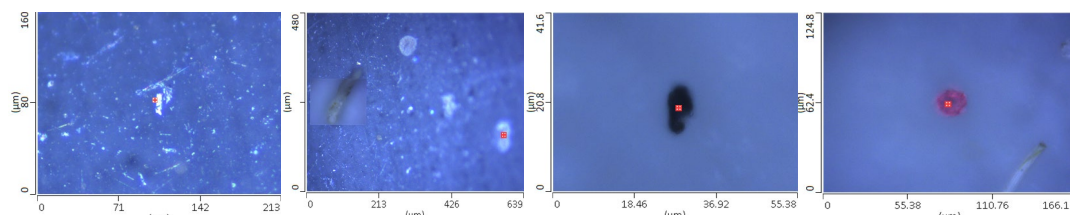


Figure 2. Particles 2, 3, 5, and 7.

Figure 1 and 2 had been obtained from OPTIR after PET bakeware had been heated at 220°C for 60 minutes. Throughout the observation, the particles underwent random inspection. Appearance, spectra, and comparative outcomes were examined for all the particles from 1 to 9. With values of 0.53, 0.76, 0.73, and 0.75, respectively, the spectra of particles 1, 4, 8, and 9 matched PET. The images showed that PET particles can come in a variety of sizes and hues, including flash white and black. Under the O-PTIR microscope, some of the particles in particles 2, 3, 5, and 7 had the same hue as PET. These particles were recognized as polyoxymethylene ($p=0.55$), HDPE (0.54), cellulose (0.72), zein (0.78), and cellulose (0.94).

In Figure 3, the whole area is presented under low magnification whereas in figure 4 the whole area was subjected under high magnification. In Figure 3, there were approximately 10 particles observed in each image except for the image D. Image D appeared to be scratched but it was identified as PET by its spectra. In Figure 4, only one particle was clearly focused in every image which results in better quality of spectra. Hence, the spectra of the particles were obtained after focusing under high magnification.

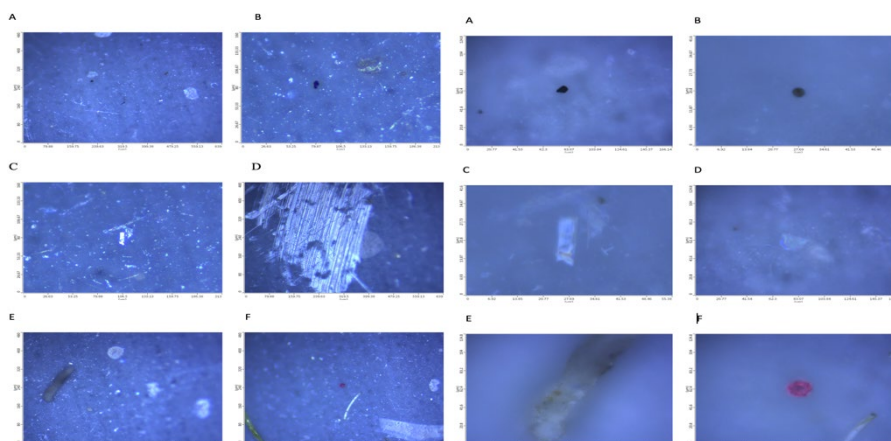


Figure 3. Image in low magnification **Figure 4.** Image in high magnification

Expected Result

After 60 minutes of treatment at 220°C, the detection of micro-PET in the PET bakeware was confirmed based on our preliminary findings. If the exposure time is shortened to 40 or 20 minutes, the likelihood of micro-PET detection would probably decline. These are our initial projections about how our project will turn out.

It can take some time to gather each particle's high-quality spectra since optimal focusing necessitates precise preparation. Despite scanning the image with feature wavelengths, our training phase experiments showed that we might also pick up particles that aren't the target ones. To ensure that only the desired particles are included in the analysis, careful attention and selection are thus necessary.

Acknowledgements

Through the Irish Research Council Pathway Programme Proposal ID 21/PATH-S/9290, Science Foundation Ireland (SFI) generously provided funding for the research detailed in this article. The researchers have been able to continue their research and expand knowledge in their domains because of this financing. This project is appreciative to the SFI-Irish Research Council Pathway Programme for their vital support of its creative and significant research endeavors.

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Orla Nic Shiurdain, BSc (Hons)

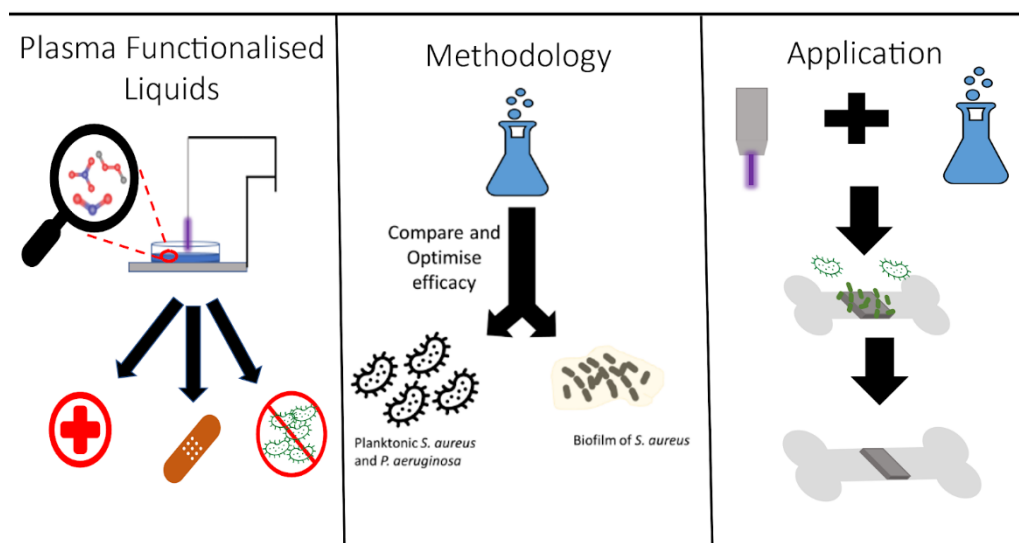
Project Title: Optimising plasma functionalised liquids for the treatment and control of orthopaedic implant infections

Project Leader: Professor Paula Bourke

Abstract

Orthopaedic implant infections are a burden on healthcare systems globally. Orthopaedic implants are commonly used to treat broken bones and to replace joints. The infection of these implants leads to longer hospital stays for patients, increased costs, and even death. Current treatments for these infections include wound debridement and treatment with antibiotics, however in some cases the implant must be removed via revision surgery. These infections are difficult to treat due to antimicrobial resistant bacteria and the formation of bacterial biofilms on the implant. Cold atmospheric plasma (CAP) and plasma functionalised liquids (PFLs) have antimicrobial and wound healing properties. These plasma treatments could be used to treat these infections. This research aims to optimise a PFL treatment to treat methicillin resistant *Staphylococcus aureus* and *Pseudomonas aeruginosa* biofilm infections which could be used in combination with direct CAP therapy. PFLs generated by four different plasma systems; the reactive species specificity system in Spark and Glow configurations, a Midiplexc system, and a submerged DBD system, were compared by comparing the PFL chemistry and the efficacy against *P. aeruginosa* and *S. aureus*. The results indicated that the chemical composition of PFLs varied between systems and that the RSS Spark and Midiplexc liquids were the most efficacious. The results also indicate that the biofilm form of *S. aureus* was more tolerant to PFL treatment than planktonic *S. aureus*. Further studies are required to investigate the toxicity of the liquids towards mammalian cells as well as the combination of PFLs and CAP treatments.

Optimising plasma functionalised liquids for the treatment and control of orthopaedic implant infections



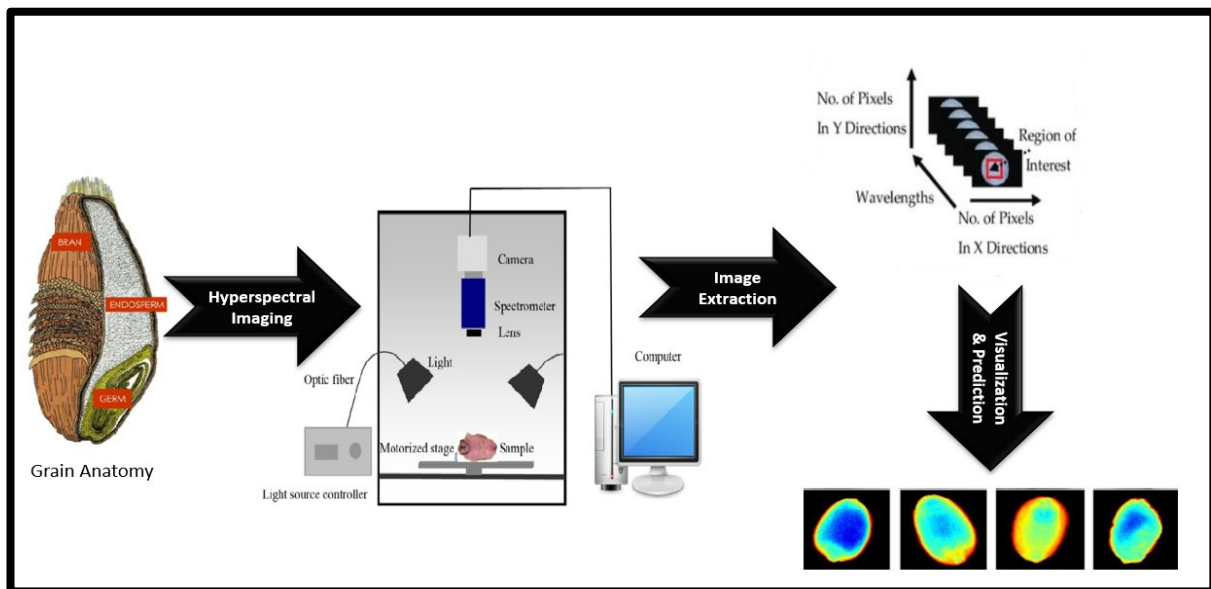
Gözde Özdoğan, BE, M.Eng.Sci., PhD

Project Title: Feasibility of spectral imaging and data fusion for classification of Turkish wheat

Project Leader: Prof. Aoife Gowen

Abstract

Wheat is the second most used grain type in the world. Identification and verification of wheat grain samples is of crucial importance because wheat grains can change in nutritional value and processability according to their origin. Moreover, verification is an obligation in seed purification. In this study, the potential of spectral imaging for the classification of wheat kernels and the effect of the data fusion via data concatenation were investigated. Spectral images of 38 different wheat varieties were collected in Vis-NIR and SWIR ranges with seeds in the crease-up and crease-down positions. Linear Discriminant Analysis (LDA) of mean seed spectra showed 93% and 75.8% classification accuracy for the samples in the up position in the Vis-NIR and SWIR wavelength range, respectively, while it was 89.5% and 81.4% for the down position. Horizontal concatenation was applied to combine wavelength ranges, and then, vertical concatenation was performed for the fusion of data in all positions which achieved the highest classification accuracy of 95.4%. This study highlights the appropriateness of spectral imaging as an objective and fast tool for the identification of wheat kernels and the promising results of data fusion to improve the classification accuracy.



Selected Recent Publications

Özdoğan, G., Lin, X., & Sun, D. W. (2021). "Rapid and noninvasive sensory analyses of food products by hyperspectral imaging: Recent application developments". *Trends in Food Science & Technology*, 111, 151-165.

ESTIMATION OF CHLOROPHYLL CONTENT IN COFFEE ARABICA LEAVES BASED ON HYPERSPECTRAL IMAGING ANALYSIS

Tina Najafpour, Aoife Gowen

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland

Abstract

The chlorophyll content of leaves is an indirect indicator of the health and nutritional status of the plant. chlorophyll content is linked directly to photosynthetic potential and primary production. An effective way to assess the Ch content is to measure reflectance, which makes it possible to assess quickly and non-destructively, *in situ*, the chlorophyll content in leaves. Hyperspectral image analysis develops a method to measure the spatial physical characteristics and spectral chemical properties and provides solutions to diagnose chlorophyll content. This study investigates the spectral characteristics of coffee arabica leaves that are kept in four different maintenance conditions based on images captured by Specim IQ and Hyspex Hyperspectral imaging systems. The study is conducted in combination with chemical Chlorophyll content determination in 100% acetone and spectral results from UV_VIS spectrophotometer. It is expected that the results from Spectral analysis provide us with the spectral signature of coffee plants, each representing different conditions, to categorize each leaf and plant in its group.

Graphical Abstract



Introduction

The chlorophyll content of leaves is an indirect indicator of the health and nutritional status of the plant. Leaves contain chlorophyll, Ch a and Ch b, as essential pigments for the conversion of light energy to stored chemical energy, and the amount of solar radiation absorbed by a leaf is a function of the photosynthetic pigment content. Thus, Ch content is linked directly to photosynthetic potential and primary production (Steele *et al* 2008).

Reflectance measurement makes it possible to assess quickly and non-destructively, *in situ*, the chlorophyll content in leaves (Gitelson *et al* 2003). Traditional methods to calculate chlorophyll content include destructive chemical extraction. While direct, this methodology is tedious and unsuitable for continuous monitoring individual plants because of its destructive manner (Liang *et al* 2017). Hyperspectral image (HSI) analyses, which combine spatial physical characteristics and spectral chemical properties, provides a potential to rapidly and non-destructively diagnose chlorophyll content to quantify photosynthesis capacity and estimate growth status (Dehua *et al* 2021). The application of reflectance spectroscopy to the estimation of leaf pigment content has recently received considerable attention (Steele *et al* 2008).

The objective of this experiment is to investigate the spectral characteristics of coffee arabica leaves in combination with chemical analysis to measure the chlorophyll content of the leaves.

Literature Overview

Since this study is in the early stages of completion, the proposed solutions will be further developed, and the following section contains an overview of previous experiments and relevant solutions.

Liu *et al* (2019) proposed a novel wavelength selection strategy using the combination of the MWPLS and GA coupled with the PLS model to select sensitive wavebands for chlorophyll content prediction. The results show that wavelength selection can remove redundant information and improve model performance.

Proshkin *et al* (2021) aimed at assessing the impact of ultraviolet radiation on main pigment content. The plant reactions were evaluated by the change in the pigment concentration (by chemical method) and by the correlation's presence between the obtained values and the vegetative indices calculated with aid of the hyperspectral images.

Song *et al* (2021) proposed a cascade method of interval-wavelength screening to reduce the data dimension and improve the crop estimation accuracy and robustness for the chlorophyll content diagnosis of maize crops. The results showed that the proposed cascaded interval-wavelength screening method could eliminate redundant and collinearity variables and improve model performances.

Narmilan *et al* (2022) provided a new framework for inferring the chlorophyll content in sugarcane crops at the canopy level using unmanned aerial vehicles (UAVs) and spectral vegetation indices processed with multiple machine learning algorithms. The findings demonstrated that the use of multispectral UAV could be utilized to estimate chlorophyll content and measure crop health status over a larger sugarcane field.

Materials and Methods

Sample Preparation

Four pots of coffee with various storage conditions will be used to prepare the coffee leaves. 10 leaves from each pot will then be chosen at random. For 42 days, the pots were placed under various conditions. Excessive irrigation, poor illumination, and frost damage. To serve as a representative of a healthy plant, one plant is maintained under ideal conditions.

Spectral data acquisition

Spectral images of the whole plant and leaves from each one will be captured using Specim IQ (Specim Ltd., Oulu, Finland), a portable spectral imaging device with CMOS spectrograph (spectral resolution of 7 nm). An LED ring light with 16" Outer Diameter, 12" Inner Diameter will be used as illumination. On the other hand, the Hypspec VNIR-1800 hyperspectral camera will be used to capture images in the range of 400 – 1000 nm. The system setup will also include a 150 W illumination system.

Chlorophyll content determination

Leaf samples are divided into small pieces and weighted to carry out chemical analysis right after the hyperspectral image acquisition. Three replications of each test are carried out. 100 mg fresh leaf tissue without veins is macerated with 10 ml of 100% acetone with a mortar and pestle to extract the pigments. The extract is centrifuged at 3000 rpm for 10 minutes. The supernatant (3 ml of each extract from each leaf sample) is then transferred into a 1-cm path cuvette. The chlorophyll content of the samples is

determined by using UV-VIS spectrophotometer (Biochrom Ltd.-Libra S22 UV/VISIBLE Spectrophotometer) with the available wavelength range of 190 to 1100 nm, and the sampling interval of less than 3 nm. The absorbance of the solution is measured at wavelengths of 662 nm and 645 nm. And the concentration of chlorophyll a and chlorophyll b is calculated according to the formulas introduced by Lichtenthaler and Wellburn as follows:

$$C_a = 11.75 \cdot A_{662} - 2.35 \cdot A_{645}$$

$$C_b = 18.61 \cdot A_{645} - 3.96 \cdot A_{662}$$

$$C_t = C_a + C_b$$

Where, the C_a and C_b are the concentrations of chlorophyll *a* and *b* respectively, and C_t is the total amount of chlorophyll (mg/L).

Expected Results

It is expected that the results from Spectral analysis provide us with the spectral signature of coffee plants, each representing different maintenance conditions, to categorize each plant in its group. Using this data coupled with the chlorophyll content from chemical experiments, water stress is expected to be analyzed.

Conclusions

Further research needs to be conducted concerning the feasibility of the suggested solutions and how they can be implemented. However, based on the evidence from existing literature, extraction of Chlorophyll content via Hyperspectral imaging is achievable. By using Specim IQ and Hyspex Hyperspectral imaging systems and analyzing the outputs, it would be possible to classify data from leaf samples into categories. And comparing the results with output data of a UV – VIS spectrophotometer can make it possible to ensure the accuracy of the experiment.

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DEVELOPMENT OF A MICROPLASTIC DETECTION FRAMEWORK USING OPTICAL PHOTOTHERMAL INFRARED SPECTROSCOPY

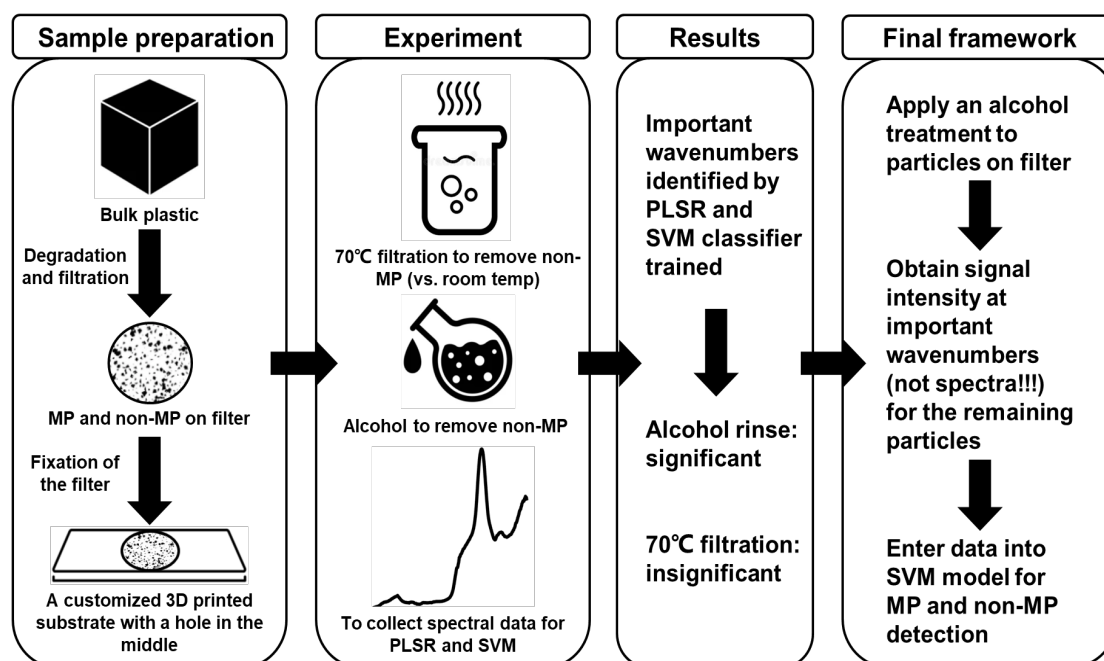
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Abstract

Evidence has shown that plastic products release microplastics (MPs), especially when exposed to external forces, such as high temperatures. However, there has not been a standardized MPs detection method. This work aims to develop an efficient and accurate framework to quantify MPs released from nylon products using state-of-the-art optical photothermal infrared (O-PTIR) spectroscopy. Firstly, MPs were released from nylon bulk and collected on filters following filtration at room temperature and 70 °C, respectively. A further treatment of the filter using alcohol was employed. A 3D printed substrate was used to accommodate the filter during the alcohol treatment and spectra collection. Hyperspectral data of MP and non-MP particles were collected and used to build a partial least squares regression (PLSR) model, from which important wavenumbers for discriminating between MPs and non-MPs can be identified. Finally, data at the identified wavenumbers were used to train a support vector machine (SVM) classifier. Results showed that wavenumbers of 1077, 1541, 1635, 1711 cm^{-1} are sufficient for discriminating between MP and non-MP particles. We also demonstrated that an alcohol treatment could significantly remove non-MP particles. There is no difference between room-temperature and high-temperature filtration. This work contributes significantly to the standardization of MPs detection methods.

Graphical Abstract



Introduction

Plastic fragments or particles less than 5mm in size are defined as microplastics (MPs). MPs have been widely present in our environment, food, and even some important organs and tissues

of the human body (Lim 2021). This is noteworthy because, currently, researchers have reported the damaging effects of MPs on biological entities. For example, Deng *et al* (2017) have demonstrated the potential for MPs to cause disruptions in energy and lipid metabolism, as well as oxidative stress. The toxicity of MPs is linked to parameters such as shape, size, and intake (Xu *et al* 2022). Studying these parameters of MPs can deepen our understanding of the harm they pose to health.

Infrared spectroscopy and Raman spectroscopy are among the most popular tools used for studying MPs (Faltynkova *et al* 2021). Using these tools (in combination with a microscope, if the particle size is small), the shape and size of MPs can be relatively easily measured; however, the quantitative analysis of MPs remains challenging. One reason is the interference from non-MP particles that have similar infrared/Raman spectra to MPs, which are often additive particles from additives used for plastic production. For example, Li *et al* (2022) reported that behenamide, a typical slip agent used in polyethylene (PE) plastic products, to be released along with the release of MP particles and it has a high degree of Raman spectral similarity with PE particles, so behenamide particles are easily counted as PE particles.

The presence of misassignment can result in inaccurate quantification of MPs, hence leading to a wrong assessment of the harm caused by them. Proper pretreatments can reduce the likelihood of misassignment. For example, Gerhard *et al* (2022) found that higher temperatures in sample preparation can remove additives such as fatty acids and their esters; Li *et al* (2022) found that alcohol is effective in removing some additives from MP samples.

Our MP samples were pretreated with alcohol and high temperatures to investigate the efficacy of these pre-treatments; an advanced optical photothermal infrared (O-PTIR) spectroscopy instrument was used to collect hyperspectral data of MPs and non-MPs; a partial least squares regression (PLSR) model and a support vector machine (SVM) classifier were developed based on the data. Ultimately, the goal of developing a reliable and accurate MPs detection framework based on O-PTIR was achieved.

Material and Methods

Sample preparation and filtration at different temperatures

Nylon bulk was degraded, and MPs were released and deposited on filters according to Xu *et al* (2021). During this step, the effects of removing non-MPs by hot and cold filtration (filtration at 70°C vs. filtration at room temperature) were explored.

Alcohol treatment

For alcohol treatment, the prepared filters were rinsed with ~ 30ml using a method that is similar to that of Li *et al* (2022).

Collection of hyperspectral data and modelling

Hyperspectral data of MPs and non-MPs on the filters were collected using the mIRage IR microscope (Photothermal Spectroscopy Corp., Santa Barbara, CA, USA). The data was firstly used for PLSR to identify the important wavenumbers that could discriminate between MPs and non-MPs. Then the data at these identified wavenumbers were used to train a SVM classifier. The sensitivity, specificity, correct classification rate (CCR) and Matthews correlation coefficient (MCC) were calculated for assessing model performance.

Data analysis

Optical images of particles and hyperspectral data were obtained using the software PTIR Studio (Photothermal Spectroscopy Corp., Santa Barbara, CA, USA, version 4.4.8075). The hyperspectral data were then masked, averaged, and compared in the MATLAB software (version R2022a), which was also the tool used for modelling. For determining significances

among different replicates, the (paired) sample t-test were carried out in Microsoft Excel or MATLAB.

Results and Discussion

Modelling performance to distinguish MP and non-MP

Based on the results of PLSR, 1711 cm^{-1} , 1635 cm^{-1} , 1541 cm^{-1} , 1077 cm^{-1} were identified as important wavenumbers. A SVM classifier was then built on spectral data at these important wavenumbers. The performance of PLSR and SVM can be described in Table 1 and Figure 1. In terms of sensitivity, specificity, CCR and MCC, both models perform well on the training set but not that well on the test set. More data are needed for improvement of the models.

Table 1. Summary of sensitivity, specificity CCR, and MCC of the built models

	PLSR (517 spectral variables)		SVM (4 spectral variables)	
	Training	Test	Training	Test
Sensitivity	0.9650	0.9433	1.0000	0.9037
Specificity	0.9667	0.9006	0.9957	0.9227
CCR	0.9658	0.9217	0.9978	0.9133
MCC	0.9316	0.8443	0.9956	0.8266

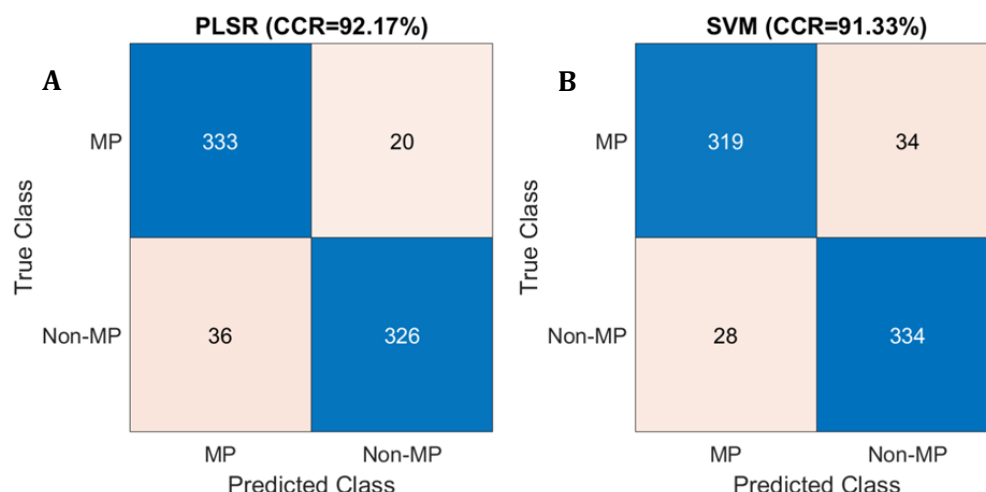


Figure 1. Confusion matrix of PLSR (A) and confusion matrix of SVM (B).

Effects of removing non-MP particles by high temperature filtration

To assess the effectiveness of particle removal, the MP particle/all particle ratio (MP/All) was used. A treatment was considered effective if it significantly increased this ratio. After particles were released from the bulk plastic, they were enriched on the filters through high-temperature filtration and room-temperature filtration, respectively. Using our developed SVM classifier, the MP/All was calculated. The MP/All from the room-temperature filtration was 0.090 ± 0.012 ; and from the high-temperature filtration was 0.08 ± 0.012 , respectively. The results of the *t*-test indicated that there was no significant difference between the data obtained from room-temperature filtration and from high-temperature filtration. This meant that, from a statistical perspective, the effectiveness of high-temperature filtration in removing non-MP was not evident. Filtration at a high temperature failed to increase the MP/Total, so it was not included in our proposed detection framework. However, high temperature filtration is not discouraged as it might work for other research experiments where the amounts of additives that could be removed by hot water are high.

Effects of removing non-MP particles by alcohol

In terms of single particles, it was apparent that some of the non-MP particles were dissolved by the alcohol treatment (Figure 2). This result is consistent with that of Li *et al* (2022). To

future explore the significance of the alcohol treatment, the MP/All was calculated using the developed SVM classifier. The MP/All before the alcohol treatment was 0.35 ± 0.16 ; and after the alcohol treatment was 0.64 ± 0.14 , respectively. The Paired *t*-test of the data indicates that an alcohol treatment of the same areas of the filter significantly increases the MP/All. In summary, an alcohol treatment is significantly effective in removing non-MP contaminants, hence, it was included in the proposed detection framework.

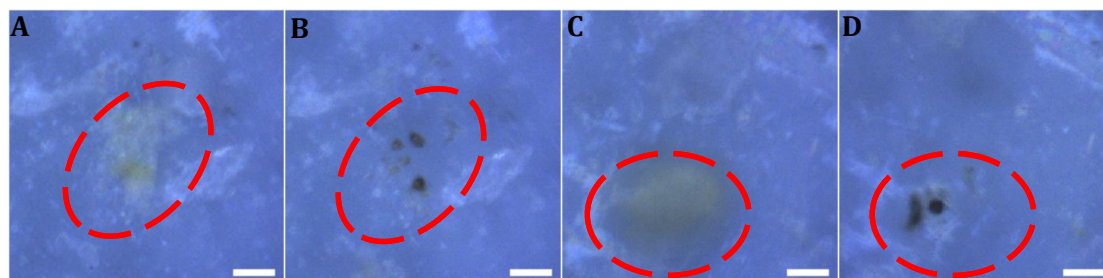


Figure 2. Particles washed away by alcohol. A and C are optical images of non-MP particles before the alcohol treatment, B and D are the optical images of the remnants after the alcohol treatment. Scale bar is 10 microns.

Conclusions

In this study, a higher temperature (70°C) did not significantly remove non-MP particles, while an alcohol treatment was significantly effective in removing non-MP particles. Our proposed MPs detection framework has been developed.

Acknowledgements

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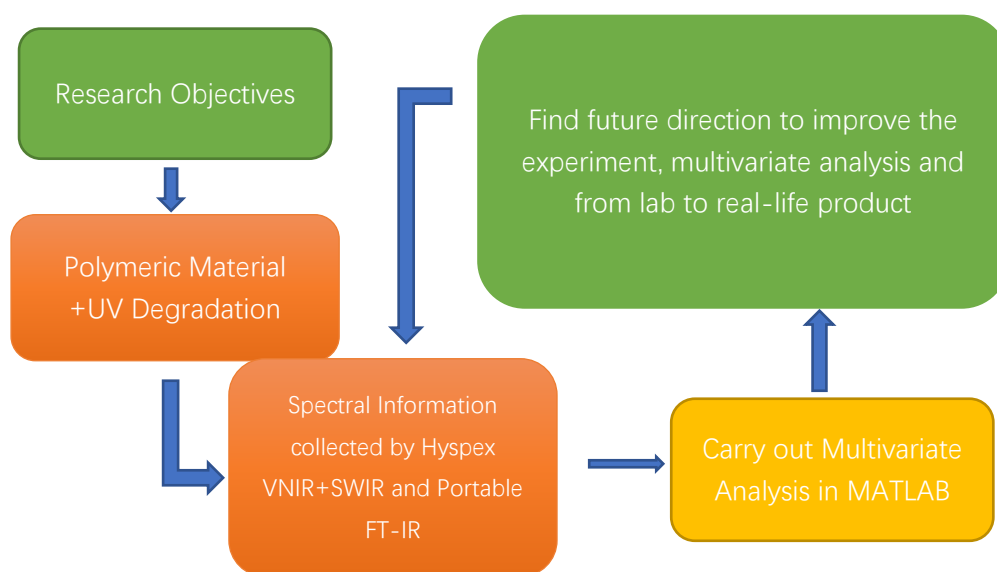
Cihang Yang, Bachelor of Science, Master of Science

Project Title: Spectral imaging for identification of polymer degradation

Project Leader: Prof. Aoife Gowen, Dr. Jun-Li Xu

Abstract

This study explores the impact of degradation (e.g., UV, Thermal) on the surface and molecular structure of polymers, through spectral imaging and vibrational spectroscopy. The main objectives were to understand these degradation effects and evaluate the potential of spectral imaging, coupled with chemometrics, for predicting or classifying changes in polymer structure due to degradation. To achieve these objectives, we employed Hypspec SWIR and VNIR cameras, alongside a portable FTIR instrument, to obtain spectral data from various polymeric samples. The collected spectral data were processed using MATLAB version 2022.b, employing multivariate data analysis, principal component analysis (PCA), and partial least squares (PLS) classification and regression modeling. The primary findings of this study indicated a capability to differentiate between different polymeric materials using the developed classification model. However, further optimization of pre-treatment methods for each material is required to enhance the model's accuracy. Moreover, extending the degradation time in future investigations would enable a more thorough observation of the complete degradation process. Incorporating more spectral data into different chemometric models could enhance the understanding of their predictive capabilities and improve their accuracy. In conclusion, spectral imaging combined with chemometrics offers promising potential for predicting and classifying degradation-induced changes in polymers, although further optimizations and investigations are needed.



AUTOMATIC RECOGNITION AND GRADING OF STRAWBERRIES USING PORTABLE HYPERSPECTRAL IMAGING CAMERA: A PRELIMINARY STUDY

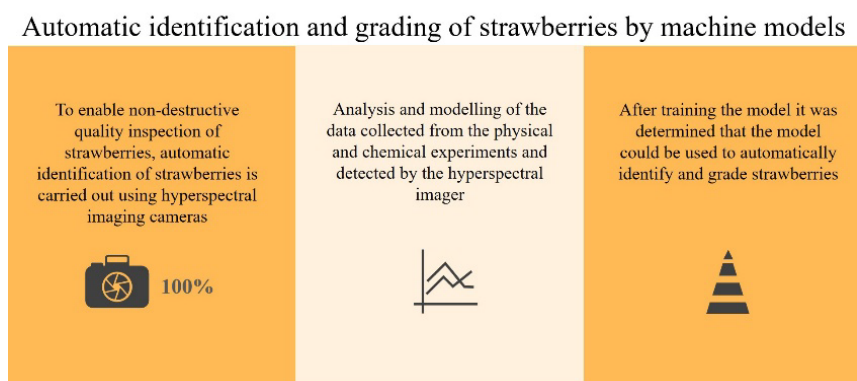
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Abstract

The nutritional benefit of strawberry is not only contributed by its vitamin supplementation and anti-inflammatory effects, but is also related to its cell renewal ability. The rapid testing of strawberries is a necessary method to check the quality. This study focuses on the analysis of four indicators including hardness, color, total soluble solids and acidity of strawberries using a portable hyperspectral imaging system and physicochemical methods. The aim is to develop a method for automatic detection of individual strawberries by deep learning, as well as to develop a grading model for strawberries by using a portable hyperspectral imaging system. The hyperspectral spectrum has a wavelength range of 397 – 1003 nm with 3.1 nm increment for a total of 204 wavelengths. Principal component analysis (PCA) was mainly applied to reduce the dimensionality of the data and to extract characteristic information from the collected data. The preliminary study indicated a positive correlation between colour and texture and the variation of spectra between different strawberry samples. In the further investigation, this study will combine the four indicators and hyperspectral imaging to develop an automatic selection, recognition and classification system for the strawberry.

Graphical Abstract



Introduction

Strawberry is famous for its vitamin composition and anti-inflammatory properties (Giampieri *et al* 2014). Recent clinical studies have confirmed that strawberries can reduce serum metabolic characteristics, improve insulin, and have viable strategies in reducing cardiometabolic dysfunction (Basu *et al* 2023). Meanwhile, with the development of technology and the improvement of living standards, people's requirements in terms of food quality are also increasing. Rapid non-destructive testing of strawberries has become a demand in the supply chain of fresh strawberries for growers or merchants and so on. Hyperspectral imaging does not require any chemical reagents and physical damage, while also detecting material changes that are not readily detectable to the human eye (Wang *et al* 2021). Therefore, hyperspectral imaging could provide some credible information for preventing spoilage of foods such as fruits.

As a result of the development and needs of the food industry, non-destructive testing and monitoring of food quality is gradually becoming a demand within the industry (Abasi *et al* 2021). Because of the local limitation of fruit testing, portable devices have become a need for industry stakeholders (Abasi *et al* 2018). Near-infrared (NIR) spectroscopy allows the monitoring of food spoilage components (Cen and He, 2007). Applicable for monitoring the physical properties such as soluble solids content in fruits. Włodarska and Mancini *et al.* have both confirmed that the soluble solids of strawberry juice can be predicted in the NIR spectral range ($R^2_p = 0.98$, RMSEP = 0.25%) (Mancini *et al* 2023; Wlodarska *et al* 2019).

The objective of this study was to develop a method for automatic detection of individual strawberries by deep learning and a grading model for strawberries using a portable hyperspectral imaging system.

Materials and Methods

Materials

Different quality of strawberries, which were purchased from of the local supermarket (Lidl, Tesco, Spar, Dunnes, Centra) and street fruit stores (Fresh Food) in Dublin, Ireland. The selection of strawberries was based on the colour and appearance of the strawberries. There are three types of strawberries: 2/3 red, all red, and a tendency to rot on the outer skin visible to the eyes.

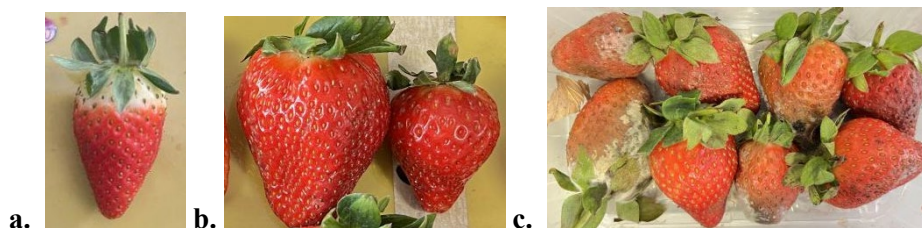


Figure 1. (a) 2/3 with red strawberries, (b) All red strawberries, (c) With rotten strawberries

Methods

Hyperspectral imaging system

The portable VIS-NIR hyperspectral system consists of three main components, the portable Specim IQ camera (Specim, Spectral Imaging Ltd., Oulu, Finland), the software system Specim IQ Studio (Specim Ltd., Oulu, Finland), and the ring light meter (Venus V29C, Guangdong Nanguang Photo&Video Systems Co., Ltd, Shantou, Guangdong, China). The working wavelength is 397-1003 nm with increment of 3.1 nm and thus 204 wavelengths in total. The sample in its packaging box was placed directly under the camera after the film was removed to obtain the hyperspectral images.



Figure 2. The portable VIS-NIR hyperspectral imaging camera

Total Soluble Solids and Colors

A portable refractometer (PAL- α , ATAGO, Tokyo, Japan) was used to measure the TSS of different parts of each strawberry. Each strawberry was divided into three parts, tip, middle and bottom, and the data of these three parts were measured and recorded. Strawberry color was

measured using a Chroma-Meter (CR-400, Konica Minolta Optical, Tokyo, Japan). Each strawberry was measured six times, and six different points were taken on each strawberry. The readings of L^* , a^* and b^* were recorded. Calculations were performed using the following formula (Lin *et al* 2019):

$$x = \frac{(a^* + 1.75 \times L^*)}{5.645 \times L^* + a^* - 3.012 \times b^*}$$

$$BI = \frac{100 \times (x - 0.31)}{0.172}$$

Titrateable Acidity and Firmness

The acidity of each strawberry was measured by grinding each strawberry homogeneously into juice and using a pH meter (520A, Thermo Orion, Waltham, Massachusetts, U.S.). The pre-experiment confirmed the feasibility of the grind-to-juice method by testing the pH value by grinding the whole strawberries into juice portions. The firmness of each strawberry was tested using a texture analyzer. Each strawberry was tested twice using a 35 mm diameter cylindrical butter at the highest point (equatorial side of the strawberry) after lying flat and at the lowest point of the strawberry growth. The maximum intensity (kg) and the positive area (kg.sec) were recorded during the test experiments.

Results and Discussion

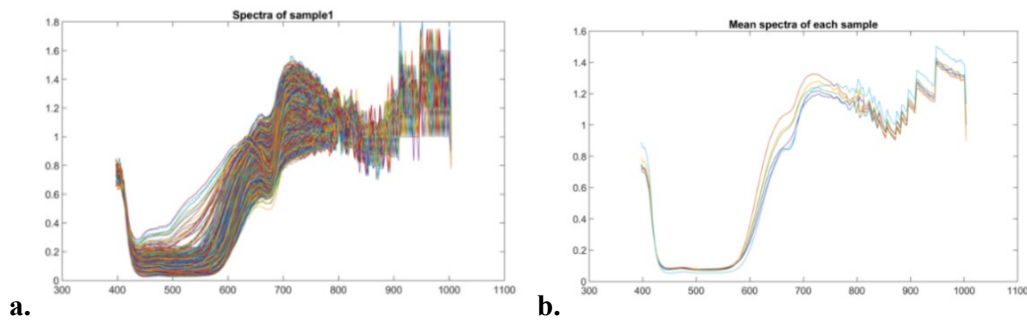


Figure 3. (a) Spectra of experimental samples, (b) Mean spectra of samples in experimental

Figure 3 shows the spectra of the strawberry samples. A number of strawberries however have different spectra, which imply the feasibility of carrying out analytical modelling.

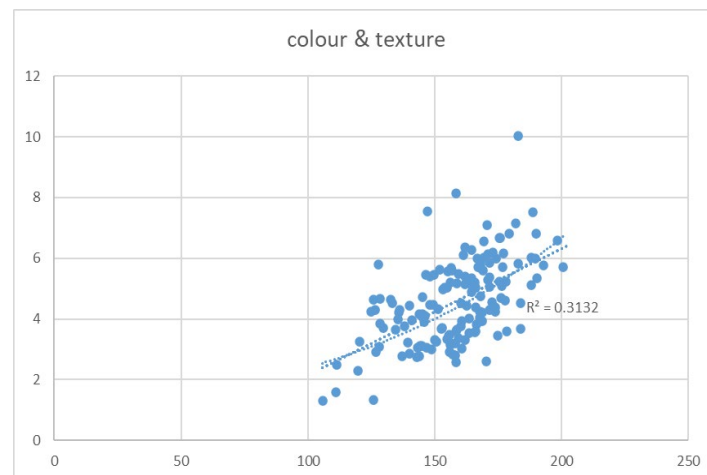


Figure 4. Correlation r value for colour and texture

Table 1. P-value for correlation between the four indicators

	colour	texture	pH	TSS
colour	1			
texture	0.56	1		
pH	-0.28	-0.19	1	
TSS	-0.065	-0.035	0.20	1

Figure 4 shows a scatter plot of the correlation between colour and firmness of strawberries. It can be seen that colour and texture show a positive correlation ($r=0.56$). Analysis the quality indication. Table 1 shows that there is some correlation between these four indicators, but not significant.

Conclusions

The full sample data has not yet been processed, after which the four quality indicators will be analyzed combined with the NIR hyperspectral data. Based on the references, it is currently envisaged that the results of the data afterwards will be able to distinguish between the different levels of ripeness of strawberries.

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PROTOCOLS FOR THE DIGITAL COLLECTION OF DATA FOR DECISION MAKING IN CROP PRODUCTION

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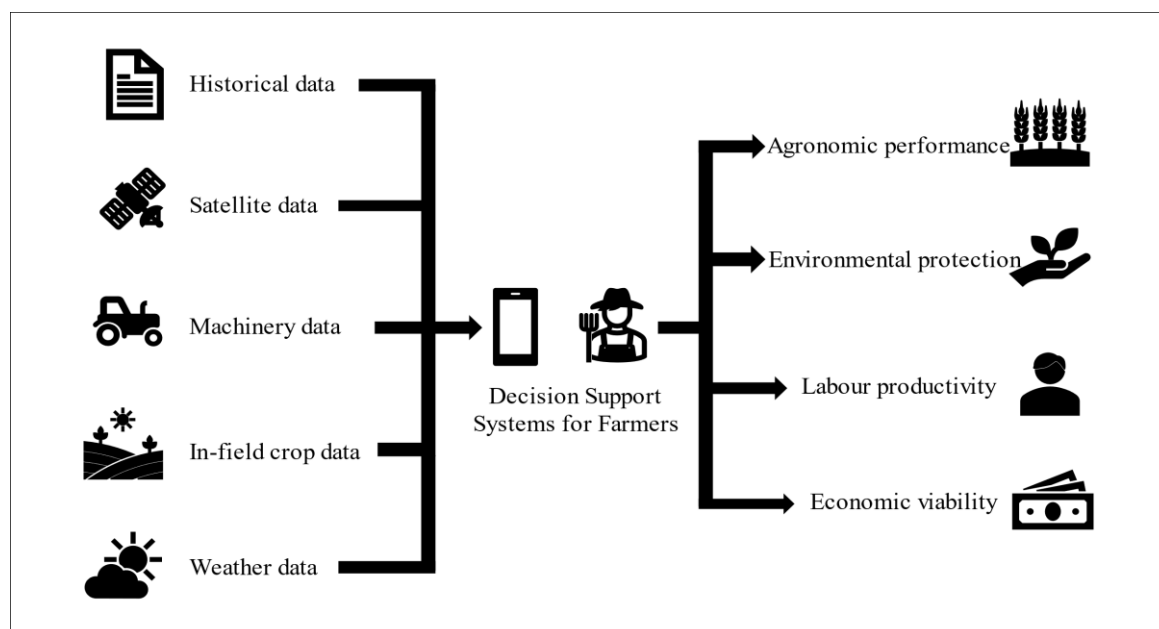
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Abstract

Digital agriculture is increasingly playing a major role in agricultural systems. As farm size increases and labour availability reduces, farmers will look to decision support systems (DSSs) to enhance their management decisions. By utilising digital collection formats, data can be integrated into DSSs to maximise farm-based efficiencies, providing accurate, real-time information to inform the decision-making process. This study will examine what protocols need to be enacted to ensure that digital data collected is accurate, precise, and consistent, increasing the reliability of suggested actions and predictions of DSSs. Digital data will be collected from various cereal crops over two years using different methods, both digitally and in an analogue form for comparison purposes and where digital collection is not possible. The data will be investigated to determine the data's accuracy, and usefulness to DSSs. This will result in a set of protocols for digital data collection in crop production and will assess the viability of utilising different digital tools for data collection for a singular DSS purpose on Irish arable farms. This will aid the development of a series of digital DSSs tools for the crop production sector.

Graphical Abstract



Introduction

Agriculture, and crop production specifically, has undergone many transformational changes in the past century as population growth, technological advances, and economic policies fuelled intensification. Mechanisation, paired with the green revolution of 1966 – 1985 of synthetic fertilisers and pesticides, increased crop yields and reduced the agricultural labour requirement. More recently, the push for decarbonisation and the mitigation of climate change has resulted

in new technologies being utilised in agriculture such as GPS, cloud-based software, Internet-of-Things, and artificial intelligence. These have been viewed as Agriculture 4.0, the fourth agricultural revolution (Rose and Chilvers 2018). These products are becoming increasingly popular, especially in crop production, as they have the potential to improve on-farm efficiencies, agronomic performance, and environmental sustainability (Birner *et al* 2021). However, most farmers are not making full use of these technologies to increase on-farm efficiencies.

As farm size increases, and labour availability reduces, farmers may be unable to manage their crops on an individual field-scale and turn to blanket treatments. This is economically and agronomically inefficient, in addition to being environmentally harmful. Decision support systems can help farmers to manage their land effectively, and to make individual decisions for individual fields. Therefore, farmers can maximise agronomic performance on an individual field basis without overapplying inputs. A higher level of management can be achieved with reduced, or at least an equivalent amount of time allocated to “walking crops” to make agronomic decisions.

Decision support systems have been used in agriculture for decades. However, while these decisions used to be manual calculations, the advancement of computer technologies led to digital tools with manual entry. While the automatic digital collection of data for use in DSSs is available commercially in some instances, it does however need to be scientifically proven that an individual farmer who uses such a tool will receive the optimal decision support for their own personal situation. DSSs have been shown to reduce herbicide use while maintaining weed control efficacy and crop yield (Sønderskov *et al* 2015), they can provide 3-16% greater profitability, 4-42% higher grain yields and up to 50% reduction in fungicide use (El Jarroudi *et al* 2015, Kettel *et al* 2022), and predict optimal seeding rate based on varietal and environmental factors (Stanley *et al* 2020). These have been proven in many areas of the world, and while there are less of these tools developed for Irish conditions, many DSSs could be potentially modified for Ireland’s temperate Atlantic climate with associated high disease pressure. These pieces of research show the multifactorial benefits to be realised from the widespread use of accurate and trustworthy DSSs. They will become especially important as Europe aims to reach its “Farm to Fork” target of reducing pesticide use by 50% by 2030.

Protocols need to be developed for farmers to be able to digitally collect data from their crops to use in DSSs. These protocols must be very simple, and not time consuming in order to maximise farmer uptake of these tools. Research on the repeatability of digital DSS predictions across a large scale involving a large number of farmers is limited. However, it has been shown that digital agriculture systems which advocate a precision agriculture system, can be “precisely inaccurate” and an “exaggerated belief” in these inaccurate digital tools can lead to a reduction in high quality management practices (Visser *et al* 2021). This is where protocols for data collection are essential to ensure farmers are using accurate data as an input into their DSS to help them make an informed decision on crop management. It is vital that these systems, which farmers believe are precise and accurate, live up to this belief to ensure no negative effects are experienced.

The objective of this study is to determine the necessary protocols for the digital collection of data for the creation of accurate decision support systems and their use by farmers in crop production.

Materials and Methods

Experimental Design

There will be three experimental trials in Year 1 at sites in UCD Lyons Farm, Kildare, Ireland. Trial site 1 and 2 will host a spring barley (*Hordeum vulgare* L. cv. Rockway) trial, while there will be spring oats (*Avena sativa* L. cv. Husky) at trial site 3. Each trial will have 4 replicate

blocks. Each plot is 3m wide. Plots in trial 1 are 30m long, while plots in trial 2 and 3 are 10m long. Trial 1 has three treatments, ploughing, minimum tillage, and no-till. These plots have been managed in their respective system since 2018 (Hobson *et al* 2022). Trial 2 has four treatments, min-till and ploughed establishment systems with and without chopped straw and additional fertiliser. Trial 3 has two treatments: min-till and plough. Each trial will be treated with commercial recommended husbandry practices (Teagasc 2015). Year 2 will involve different crops at the same trial sites, with crops to be chosen based upon standard rotational practices.

Data Collection Completed

Pre-sowing, soil sampling was carried out to determine total organic carbon and bulk density. A Eijkelkamp® soil corer with a diameter of 50mm and 300mm length was used for bulk density, while a Eijkelkamp® gouge corer with a diameter of 30mm was used to sample to 300mm for total organic carbon. Each soil core was split into 0-10cm, 10-20cm, and 20-30cm in field. For bulk density, fresh samples were then weighed prior to drying. Samples were placed in an oven for 24h at 105°C and reweighed to determine moisture % and thus dry bulk density (Campbell and Henshall 2001). Total organic carbon samples were sieved to 2mm, and dried, to be analysed via a total C analyser at a later date. These datasets are helpful for comparing selected treatments, and to make decisions regarding soil intensity and an indicator of soil health.

Data Collection Proposed

Other data to be collected includes establishment rates, plant counts, solar radiation interception, aboveground biomass, leaf area index, leaf wetness, disease incidence levels, grain yield and quality. Where possible, these will be collected continuously (e.g. leaf wetness) and transmitted via LORAN C network to the farm office. Where collection tools are not connected to this network, or are not located within the field full time, data will be collected at key timings throughout the year where management decisions need to be made. Weather data will also be collected, including precipitation, soil moisture, soil temperature, and air temperature. Normalised Difference Vegetation Index (NDVI) will also be collected via satellite, in addition to operational data from agricultural vehicles and machinery via the Trimble Pro ® management software which records inputs applied, time and date of application, and operating overheads such as diesel. The in-field crop related data will be collected in a digital and analogue format where possible.

Data Analysis

Data obtained from digital and analogue collection will be analysed for accuracy and compared via root mean square error (RMSE), relative standard error (RSE), and concordance correlation coefficient (CCC). This will determine the accuracy of digital collection to standard physical collection. It will determine which datasets can be collected via digital tools. The different treatments of trial 1, 2, and 3 will be statistically analysed via ANOVA with a significance level of 95% ($p < 0.05$). This will help to determine if different protocols are necessary for different crop establishment systems that an Irish arable farmer may use.

Expected Results and Discussion

This study will result in a set of protocols for the accurate and consistent digital collection of crop production data. Where a protocol cannot be created, a general methodology for the creation of a protocol will be created. This will allow for the harmonisation and homogenisation of datasets with data collected with different software packages and digital hardware which may be operated by any individual farmer. This research will determine the accuracy of this digital data collection compared to analogue collection where possible. It is anticipated that digital collection will be as accurate, if not more accurate than some analogue forms of data collection. It is expected that these protocols will be accurate for different crops, at different stages of growth, and in different location-based factors such as climate and soil. Where digital

collection is deemed unfit for purpose, novel methods of collection of that particular dataset will be investigated.

Expected Conclusions

This study will examine the protocols required for the digital collection of data to ensure the accuracy of decision support systems based on these protocols. It will show that digitally collected data can be accurate and increase on-farm efficiencies. This will allow for decision support systems to be accurately calibrated and operated with digitally collected data, providing fast and simple decision support for farmers. This project will trial these protocols and collection methods through the creation of a series of simple, farmer-friendly decision support systems.

Acknowledgements

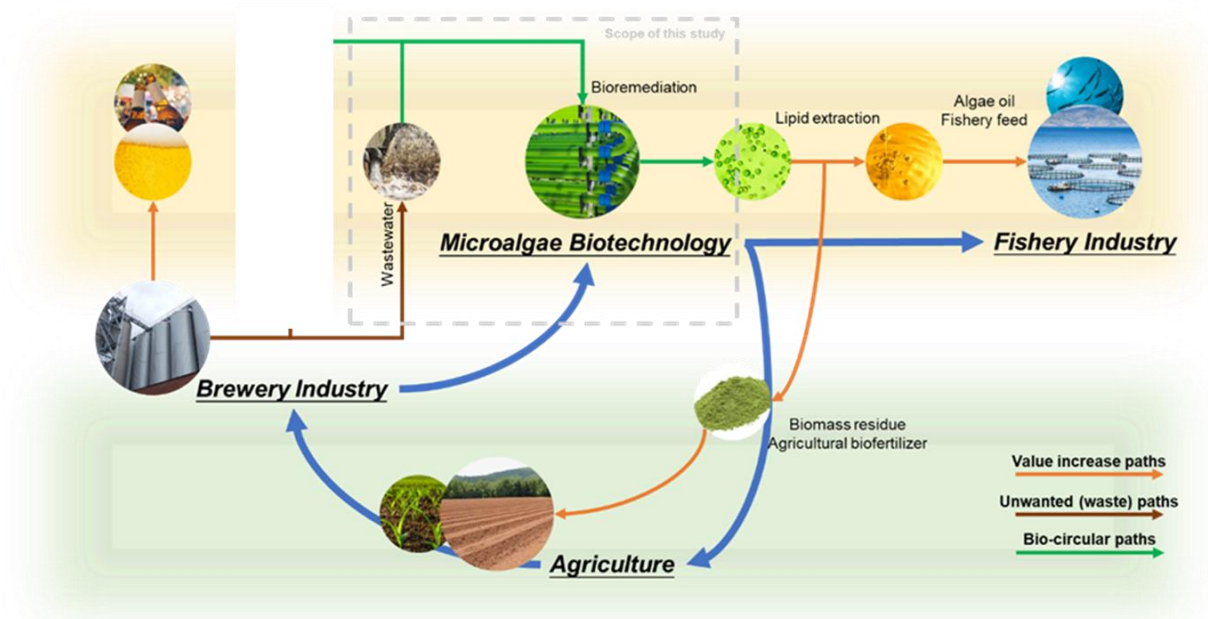
This research is being undertaken as part of the DIVINE project (Demonstrating Value of agri data sharing for boosting data Economy in agriculture), funded by the Horizon Europe programme of the European Union under Grant Agreement ID 101060884.

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Abstract

An effective and green technology for wastewater treatment is microalgae bioremediation. With their natural advantage to absorb dissolved nutrients in water, microalgae can perform valorisation of chemicals from wastewater, thereby accumulating biomass for animal feed and biofertilizer. Microalgae also has the potential to adapt to many types of wastewaters. However, wastewater characteristics such as chemical concentration, pH, and salinity could vary significantly. Therefore, some wastewater profiles may limit microalgae growth. This study aims to investigate the brewery wastewater's impacts on microalgae growth and their bioremediation ability. Within the 14-day cultivation in contrast with the growth on respective standard media, the brewery wastewater boosted *C. vulgaris* growth by 63%, while depressing the *N. oceanica* growth by 66%. Both microalgae have achieved significant nutrient removal of the wastewater.

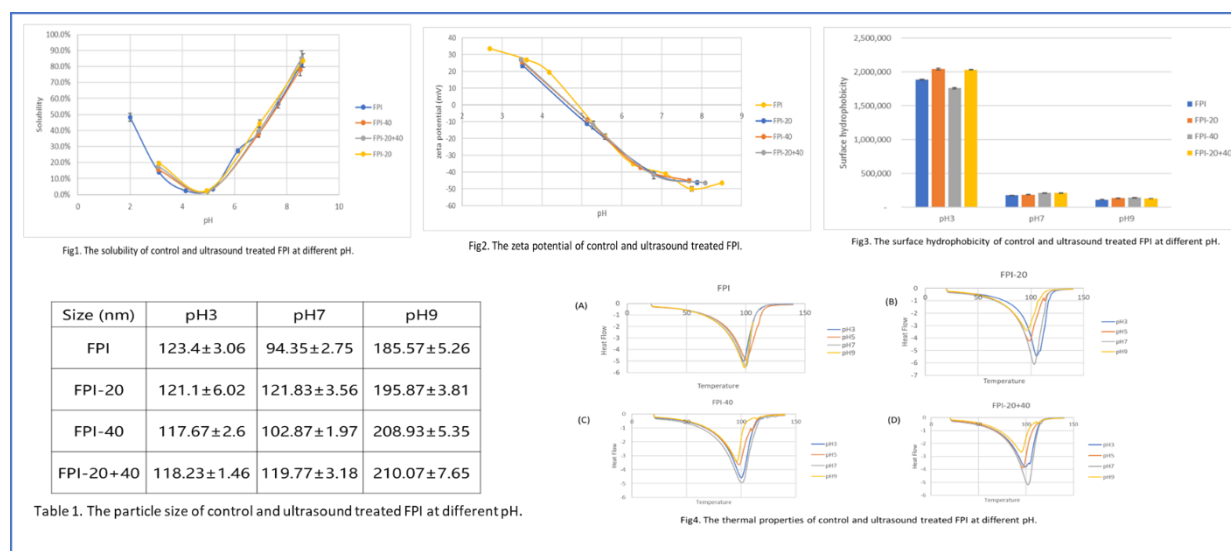


Project Title: The effects of ultrasound treatment with different frequency on physicochemical and functional properties of fava bean proteins

Project Leader: Prof. Da-Wen Sun, Prof. Song Miao

Abstract

Fava bean proteins (FPI) are candidates for animal protein alternatives. However, the functional properties of them meet many challenges. It is necessary to use modification approaches to improve functional properties and change the limitation. Ultrasound treatment is an efficient modification method which can change the protein particle size distribution and functional properties. The research is to evaluate the effects of ultrasound treatment with different frequency on physicochemical and functional properties of fava bean proteins. In the project, fava bean protein isolates (FPIs) were treated by ultrasound with different frequency (20 kHz, 40 kHz and 20+40 kHz). Then the solubility, zeta potential, particle size, surface hydrophobicity, and thermal properties of treated and untreated FPIs were researched. The treated FPI had higher solubility than the control FPI, especially at the pH below pI. Treated FPI had higher surface hydrophobicity than untreated FPI. The particle size of treated FPI was larger than that of untreated FPI at pH7 and pH9, while was smaller than that of untreated FPI at pH3. Treated proteins had a decrease of the enthalpy of protein denaturation compared to untreated proteins. Overall, ultrasound treatment changed the physicochemical and functional properties of FPIs, which will benefit its application. Ultrasound treatment with 20 kHz had more influence in the properties compared to that with 40 kHz and 20+40 kHz.

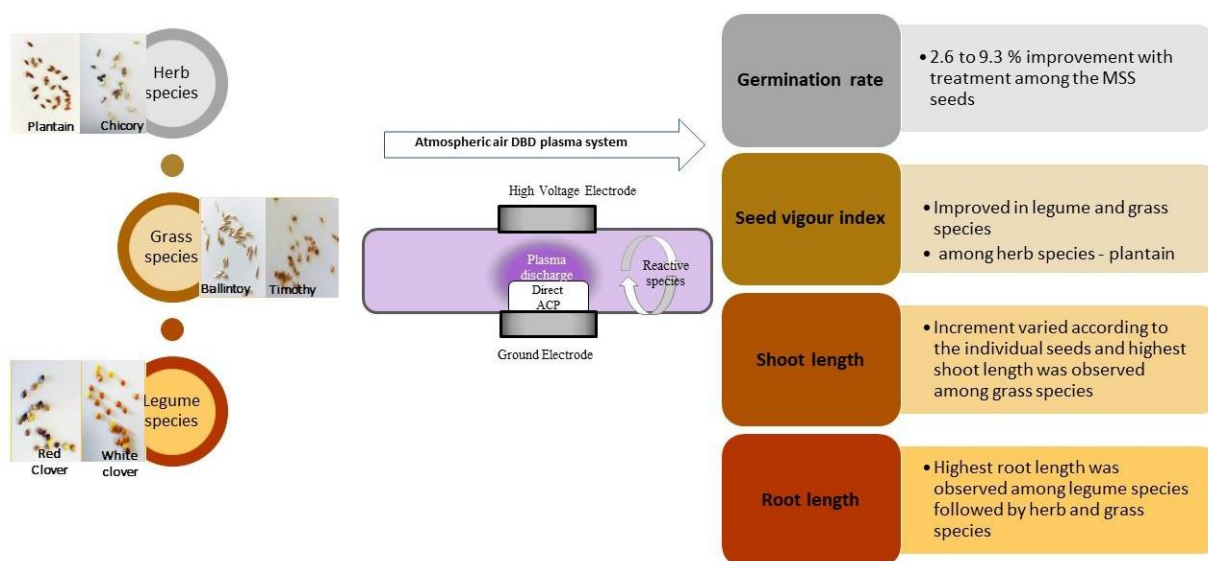


Project title: AgriPlasma – Non-thermal air plasma treatment of multispecies swards seeds for reduction of greenhouse gas emissions

Project Leader: Prof. Paula Bourke

Abstract

The maintenance of grasslands for animal forage is essential in countries with large ruminant livestock herds. Many grasslands are mono sward and require a high chemical fertilizer input for optimum yield. The agricultural sector seeks alternative approaches to reduce the need for and reliance on artificial Nitrogen application and that may also reduce greenhouse gas (GHG) emissions. Incorporating diversified and multi-species swards (MSS) in the grasslands for ruminant forage can reduce GHG emissions and improve ruminant health. Moreover, MSS with diversified root systems can enhance soil nutrition. However, despite the potential advantages presented though use of MSS, the establishment and maintenance of MSS grasslands is challenging. The AgriPlasma study focuses on developing a green and sustainable seed priming technology to address limitations and enhance the performance of multi sward species comprised of selected grass, herbs, and legume seeds. The current study examines the responses of grass, herb and legume seeds to cold plasma seed priming treatments. An atmospheric Dielectric Barrier Discharge (DBD) reactor using air as its input gas was employed. The plasma process parameters of treatment duration (10 and 30 s) and input voltage (50, 60 and 70 kV) were assessed for the seed treatment. Thereafter, treated seeds were germinated in the controlled conditions by maintaining 25 °C and 12 hr light cycle. The seeds were analysed for germination rate, seed vigour index (SVI), shoot and root length. Each seed type responded differently. The enhancement in the seed germination rate and SVI was observed across the species. These findings suggest that plasma has potential for use as a seed priming technology and that seed type and structure are important factors.



ECONOMIC AND FEASIBILITY ANALYSIS OF RENEWABLE ENERGY INSTALLATION IN MEDIUM-SCALE DISTILLERIES

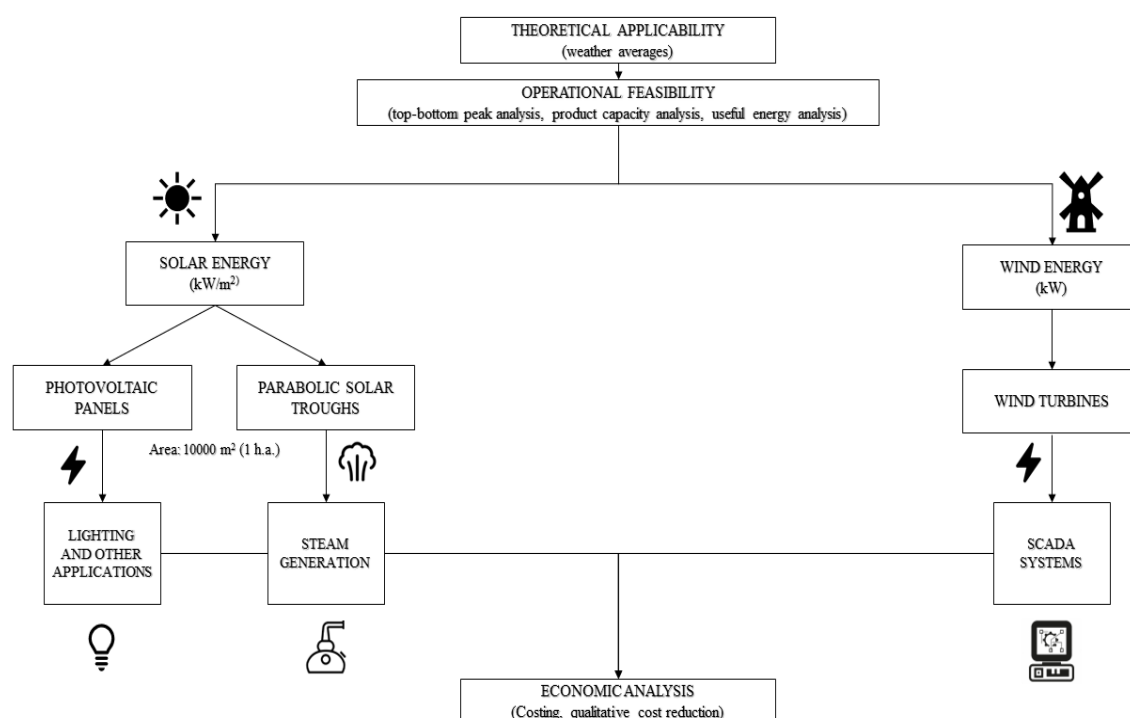
Rahul Macharaja, Patrick Grace

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

For a sector as robust and emerging as the Irish alcoholic beverage industry, sustainability is of prime importance, and strategies towards the same would begin with exploring possibilities of renewable energy installation. As such, this study is aimed at assessing how solar and wind energy could be used to power certain operations in a medium-scale whiskey distillery, which might be vulnerably placed in what one could call a balance between operational and economic feasibility. Taking as a model a distillery in the Southeast region of Ireland, historical weather data is studied, and eventually, the feasibility of installing photovoltaic panels and wind turbines for electricity, and parabolic troughs for heating, is determined. Furthermore, the expenditure is also studied to assess the qualitative driving factors of cost reduction.

Graphical Abstract



Introduction

As of 2022, Ireland became the base for over 40 distilleries, with as much as 100 million litres of pure alcohol (LPA) annually, with over 3.5 million casks filled to generate the final alcoholic product (Irish Whiskey Association 2022). With such an industry having achieved a sales growth of 150% over the last 12 years, reducing carbon footprint must be a major consideration in view of the sustainable development goals (SDGs). That said, renewable energy sources as alternatives to run distillery operations shall be, when/if successful, significant progress towards Scope 1 of carbon footprint reduction. Keeping this in mind, this study seeks to assess the

possibilities of installing equipment to harness solar and wind energy – but with specific focus on medium-scale industries.

The question as to why medium-scale industries are considered may be answered by a hypothetical understanding that medium-scale industries require a challenging balance to be struck between operation and costs. If the model of a medium-scale whiskey distillery is taken up (as in the case of this thesis), two major types of energy-intensive operations must be looked at – one, the heat-intensive mash conversion and distillation processes that require about 1.6 million kWh of steam every year, and the electricity-intensive automation that requires several panels of Programmable Logic Controllers (PLCs) to run the Supervisory Control and Data Acquisition (SCADA) systems, consuming 1.5 million kWh of energy annually (these values have been taken from the distillery studied as the model; the name is not disclosed for the sake of confidentiality).

Having known the energy requirements, the next point of observation would be how renewable energy could play a role in replacing, or alleviating the dependence on, conventional energy sources. For this, two types of renewable energy – solar and wind, are studied. Consequently, three types of equipment are also studied – photovoltaic solar panels to provide power for on-site lighting and domestic appliances, parabolic solar troughs (constructed sets of reflecting collectors) to transfer heat for generating steam for distillery operations, and wind turbines to convert mechanical energy into electricity for the automation systems.

Taking note of the fact that the driving factor for solar and wind energy is, in fact, the weather in the surrounding region, a theoretical value of average power output from the extent of prevalence of sun and wind could be obtained. However, confidence of practicality may be hard to establish based on average weather data values alone. For practical satisfaction, hourly or daily data may prove to be more precise to actually conclude whether or not it would be a good initiative to shift the reliance on energy to renewable sources, partly or completely. Finally, from the perspective of economic satisfaction, capital and operational expenditure, cost reduction, and assessment of reasonableness of renewable energy is crucial in this study.

Materials and Methods

The methodology for this study is kept simple yet detailed, with an aim to achieve intrinsic results that ultimately point to the yes-or-no factor of feasibility. As such, the steps of working are broken down in a top-to-bottom approach.

Theoretical applicability – weather averages

Historical data is collected from two databases – Met Éireann and Weather Underground, for the past four years (2022 to 2019). Parameters considered include global irradiation (in Joules/cm²), the average sunshine hours per month, average wind speeds (in km/h), and net precipitation (in mm) to better understand sunny months in a year. Thereafter, the theoretical solar power (in kW/m²) and wind power (in kW) are calculated for the monthly and yearly averages.

Operational feasibility – equipment-based actuals

To improve the accuracy and precision to the maximum extent possible on a data analysis level, hourly and/or daily values of solar irradiation and wind speeds are collected from the same databases, and these daily values compared to the minimum irradiation or wind speeds required to run the panels, troughs and turbines at their rated capacity. With this, the actual power output is calculated and the deviation from the theoretical data observed – to prove the necessity of more intricate data observation. To avoid redundancy, a top-bottom peak analysis is carried out – only the most and least sunny/windy months of a year are taken into account and studied as two extremes, the deviations of which are duly noted.

Economic feasibility

To understand the economic aspects, the study aims to estimate the capital expenditure (CAPEX) costs and carry out a model risk assessment to establish possible operational cost factors, while also understanding the driving factors of cost reduction using the experience curve theory (Steffen *et al* 2020). Keeping in mind the limitations associated with theoretically assessing costs, the economic analysis is kept as qualitative as possible to offer better clarity instead.

Results and Discussion

Acquired results of theoretical feasibility

For the theoretical feasibility power output calculations, efficiencies were taken at 20% for photovoltaic cells (University of Michigan 2021), at 75% for parabolic troughs (Kutscher *et al* 2012), and at a capacity factor of 25% for wind turbines (IEA-Wind 2021). The output values were calculated for a set area of 1 hectare (10000 m²).

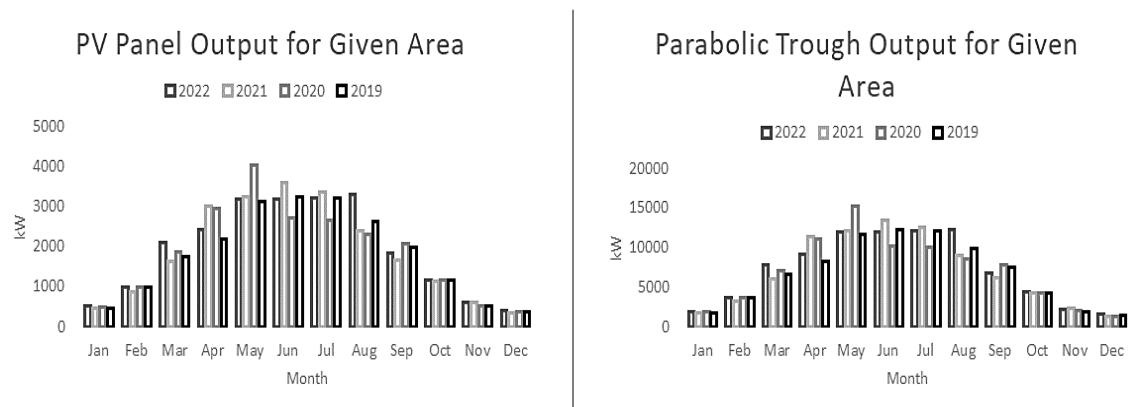


Figure 1. Theoretical feasibility for photovoltaic (PV) panels (left), and parabolic troughs (right)

Expected results from operational and economic feasibility studies

Keeping in mind the drawbacks of theoretical estimations, it is expected that the operational feasibility shall be less promising in numbers, but more precise and hence a good indicator of how beneficial renewable energy installation might be. Based on the efficiencies and capacity factors, this shall be determined for the top and bottom peaks. In terms of the capital expenditure and risk assessment of operational expenditures, clarity in determination of cost-cutting factors is anticipated. On the whole, a possibility of applying solar and wind energy as a reduction in the burden of non-renewable energy sources is a conclusion this study will arrive at.

Conclusion

In summation, this study serves as a primitive yet considerably important method to carry out a preliminary analysis of renewable energy feasibility in specific industries of specific scale, which is the very idea of the work done – to establish confidence purely with available data, and to potentially help industries reconsider the requirement of directly implementing trial operations or pilot models for analysing feasibility.

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EFFECTS OF FEED ADDITIVES ON AMMONIA AND GREENHOUSE GAS EMISSIONS DURING MANURE STORAGE

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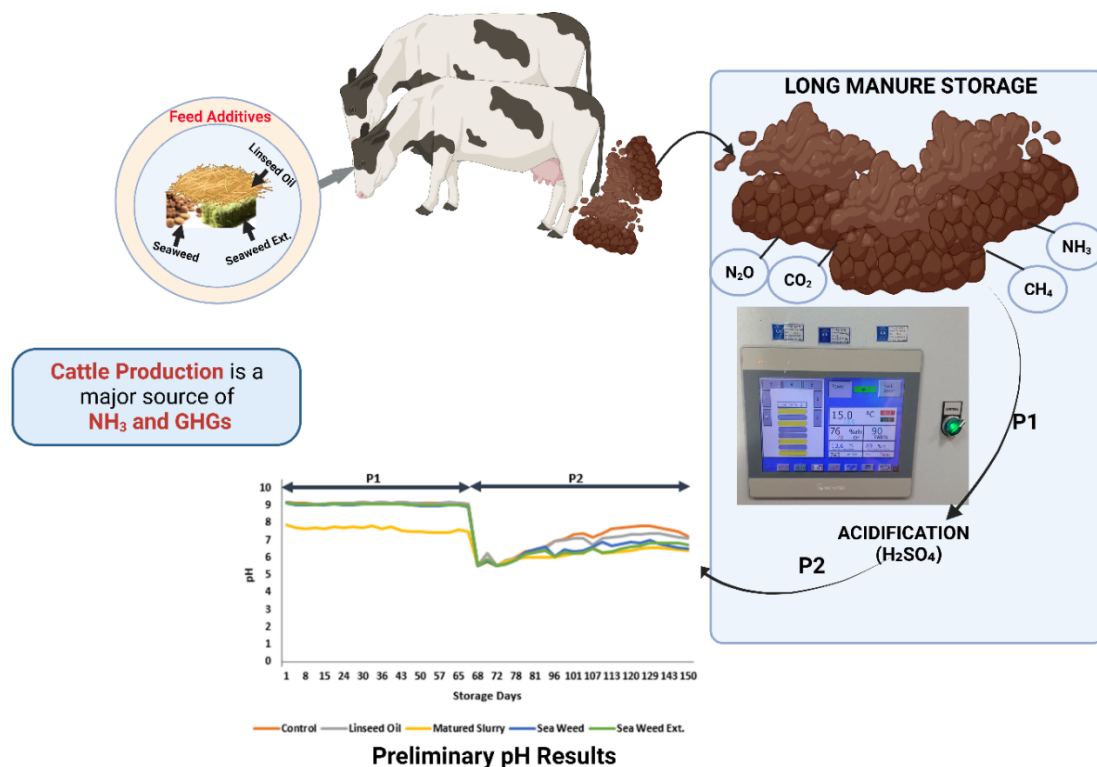
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Abstract

Ammonia (NH₃) and greenhouse gases (GHGs) are major environmental concerns from cattle production. Manure management accounts for over 80% of NH₃ emissions in Ireland (Buckley *et al.*, 2020). Several dietary manipulations, especially the use of feed additives (FAs), have been adopted to mitigate gaseous emissions from cattle during housing. However, there is limited information on the effects of these FAs when the resultant manure is stored for long periods. This research investigated the effects of the following feed additives: linseed oil (LIN), seaweed (SW) and seaweed extract (SW Ext) on NH₃ and GHG emissions during manure storage. After 68 days, treatments were acidified to pH 5.5 using sulphuric acid to determine the additive + acidification effect on gaseous emissions. NH₃ was sampled using a dynamic chamber technique with a photoacoustic gas analyser while the GHG emissions were sampled in a static chamber and measured using gas chromatography. Preliminary results show that the FAs decreased slurry pH (after acidification), a major determinant of emissions during storage. At the end of the storage period, LIN, SW and SW Ext reduced pH by 1.3%, 10% and 6.6% respectively. The results for the gaseous emissions are awaiting further analysis.



Introduction

Manure management is an important source of greenhouse gases (GHG), especially methane (CH_4) and nitrous oxide (N_2O) and air pollutants such as ammonia (NH_3) in livestock production systems. Agriculture, specifically livestock production, accounts for over 80% and 60% of the total global NH_3 emissions, respectively (Behera *et al* 2013). Globally, these gaseous emissions need to be reduced if environmental targets are to be met. For instance, the National Emissions Ceilings Directive requires Ireland to reduce NH_3 emissions by 5% in 2030 when compared to 2005 levels (European Environment Agency). Feeds and feed additives can determine the composition and quantity of manure produced, thereby controlling what enters the manure management chain (MMC). Additives such as seaweeds (Glasson *et al* 2022) and linseed oil (Doreau *et al* 2018) have reduced enteric CH_4 emissions when added to cattle diets at the housing stage, without negative impacts on productivity. While this is positive, emissions during other phases of the MMC (storage and land spreading) are a significant source of gaseous emissions. However, limited studies extend gaseous measurements from feed additives to the manure storage stage. Hence, there is limited knowledge of the downstream effects of feed additives on emissions when manure is stored.

This research aims to examine the effects of feed additives (seaweed and linseed oil) on NH_3 and GHG emissions from cattle manure during storage.

Materials and Methods

Urine and faeces were collected from the animals during a previous feeding trial. Briefly, 16 animals were divided into four groups and were fed four individual treatments— Control (CON), Linseed oil (LIN), Seaweed (SW) and Seaweed Extract (SW Ext). The animals were fed a 60:40 forage to concentrate diet. The additives were added at 4% (LIN) and 2% (SW and SW Ext) of the overall diet. *Ascophyllum nodosum* (brown seaweed) was the seaweed used, but the phlorotannins were condensed in the SW Ext. treatment. The faeces and urine of each treatment were mixed and sieved to form a homogenous slurry in a ratio that produced a dry matter (DM) of 6% (Bourdin *et al* 2014). Slurry from an underground storage tank in the beef farm of Teagasc Johnstown Castle, henceforth called - Matured Slurry (MS), was collected and formed the fifth treatment in this experiment. Unlike the first four treatments, which were purely faeces and urine, the MS treatment contained wash water, waste feed and other farming wastes, which is typical of slurry on commercial farms. This also meant that the MS treatment had a lower pH at the start of the experiment. Sub-samples were collected before the experiment for slurry characteristics analysis. 1.6 kg of the mixed slurry was then transferred to a 2 L-capacity urine container for a laboratory-scale incubation in a temperature-controlled growth chamber. The temperature and relative humidity of the chamber was set to 15°C and 76% respectively. Each treatment was replicated five times in a randomised block design. To simulate air movements during manure storage in a slatted shed, 10 holes were drilled into the lids of the 2 L containers. The total storage period was 150 days but was divided into two phases, one and two. In phase one (P1) (first 68 days) samples were stored without acidification to determine additive-only effects on emissions. In phase two (P2) of the experiment (post 68th day), all treatments were acidified with sulphuric acid to pH 5.5 to determine the effects of additive + acidification on emissions during manure storage.

Ammonia and GHG emissions were sampled three times per week at the start of the experiment and then twice per week thereafter. Ammonia was sampled using a dynamic chamber technique with an INNOVA 1412i photoacoustic gas analyser while the GHG emissions were sampled with a static chamber technique as described by (Kavanagh *et al* 2019, Connolly *et al* 2023), collected gas will then be analysed using a gas chromatograph. pH and temperature readings were also taken every sampling day.

Data Analysis

R programming will be used for all statistical analysis. A one-way ANOVA using each treatment as an independent variable will be carried out at the end of the experiment, after which a Shapiro-Wilk test will be used to assess possible statistical differences.

Preliminary Results and Discussion

Gaseous Emissions

The results for the gaseous emissions (NH_3 and GHGs) are not yet available. Raw NH_3 results have not been subjected to data analysis as stated above while the GHG samples are awaiting analysis.

Slurry pH

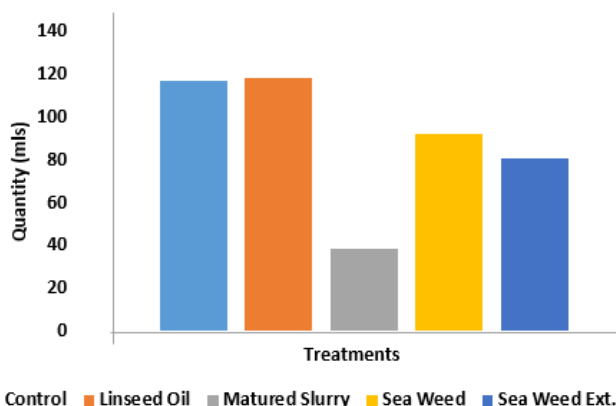


Figure 1. Quantity of Sulphuric Acid added to begin Phase 2 (P2) of the slurry storage.

Compared to the control, the seaweed treatments required less acid to arrive at a pH of 5.5 (Figure 1). Slurry acidification during storage is an established recommendation for gaseous mitigation (Misselbrook *et al* 2016). This may provide an economical advantage to commercial farms that adopt acidification during manure storage.

Preliminary results shown in Figure 2 demonstrate there was no difference in pH among treatments during P1 of the experiment. However, pH differences were observed in P2. At the end of P2, LIN, SW and SW Ext reduced pH by 1.3%, 10% and 6.6% respectively when compared to the control. Treatments did not return to original pH post acidification. Further analysis will be carried out to ascertain significance ($p < 0.05$).

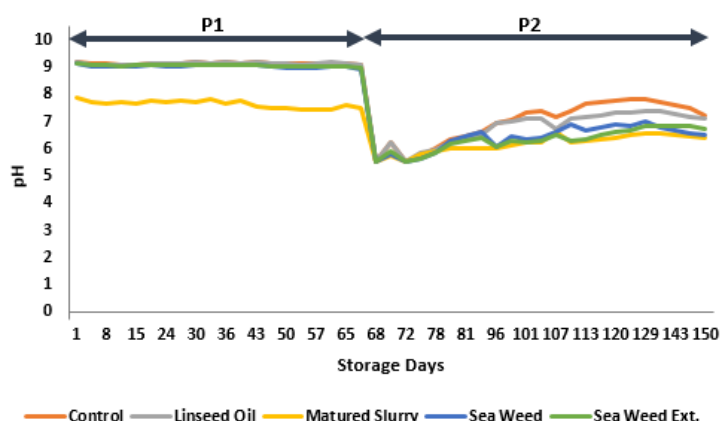


Figure 2. Preliminary pH results showing the effects of feed additives (FAs) alone (P1) and FAs + acidification (P2) on pH during slurry storage.

Slurry pH is a major factor in gaseous emissions. NH_3 emissions, for instance, increase with increased slurry pH during storage, and a reduction in NH_3 emissions by up to 80% can be achieved when slurry pH is decreased to 5.5 (Hafner *et al* 2019). Although the CH_4 results are not yet ready, acidification has been reported to reduce CH_4 emissions during manure storage (Petersen *et al* 2012, Overmeyer *et al* 2023).

Conclusions

Feed additives used in cattle production can contribute immensely to meeting environmental targets from agriculture. FAs can affect the chemical composition of manure thus influencing gaseous emissions. While some FAs have reduced gaseous emissions during cattle housing, studies need to be extended to their effects on manure storage to ensure observed positives are continuous in the manure management chain. The FAs in this study reduced slurry pH during the P2 storage period and this is likely to affect gaseous emissions.

Acknowledgements

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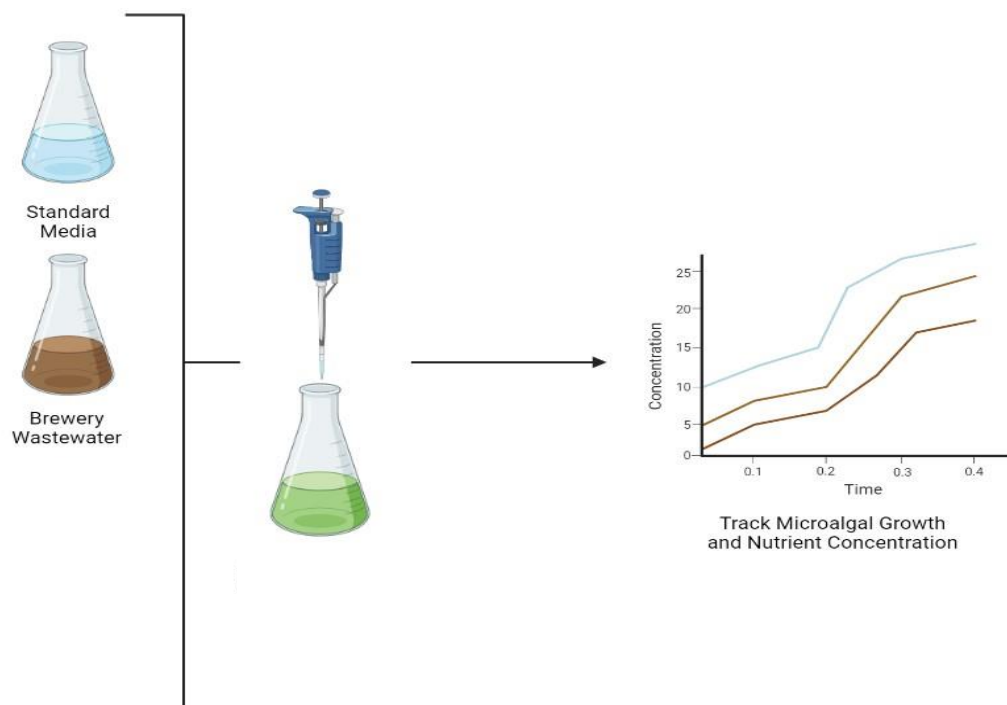
Sari Nuwayhid, BE

Project Title: Cultivation of Microalgae on Brewery Waste

Project Leader: Dr Ronald Halim

Abstract

Brewery wastewater (BWW) is an industrial waste with the potential to be used as a culture media for cultivation of microalgae because it is rich in nitrate and phosphate which are essential nutrients for algal growth. Three microalgae cultures were prepared in duplicate using culture medias of standard media and BWW. The cultures are prepared using 95 mL of cultivation media and inoculated with *Nannochloropsis* to an optical density (OD) of 0.1. Samples are taken every other day to compare the microalgae growth rate and nutrient uptake from the media over a two-week cultivation cycle. The cultivations are used to determine whether *Nannochloropsis* can treat the BWW and render these waste streams fit for disposal to the environment.

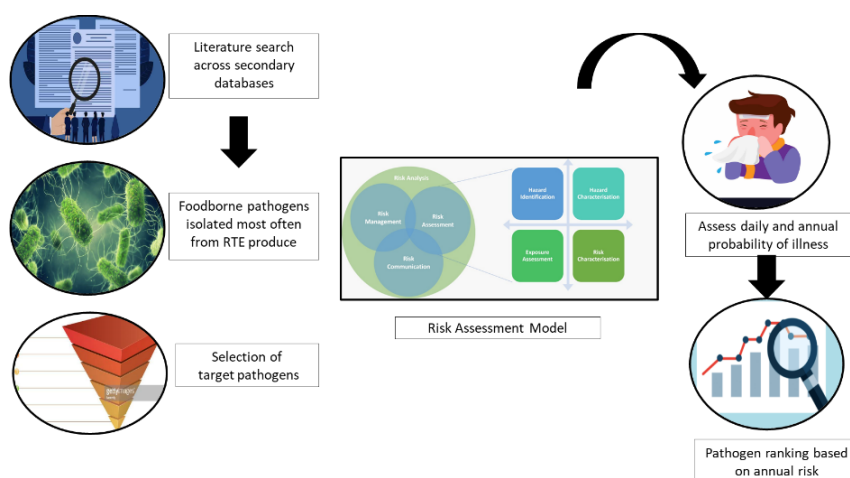


Project Title: Ranking strategy for microbial hazards associated with RTE fresh produce following the use of recycled wastewater for irrigation

Project Leader: Prof Enda Cummins, Dr Rajat Nag

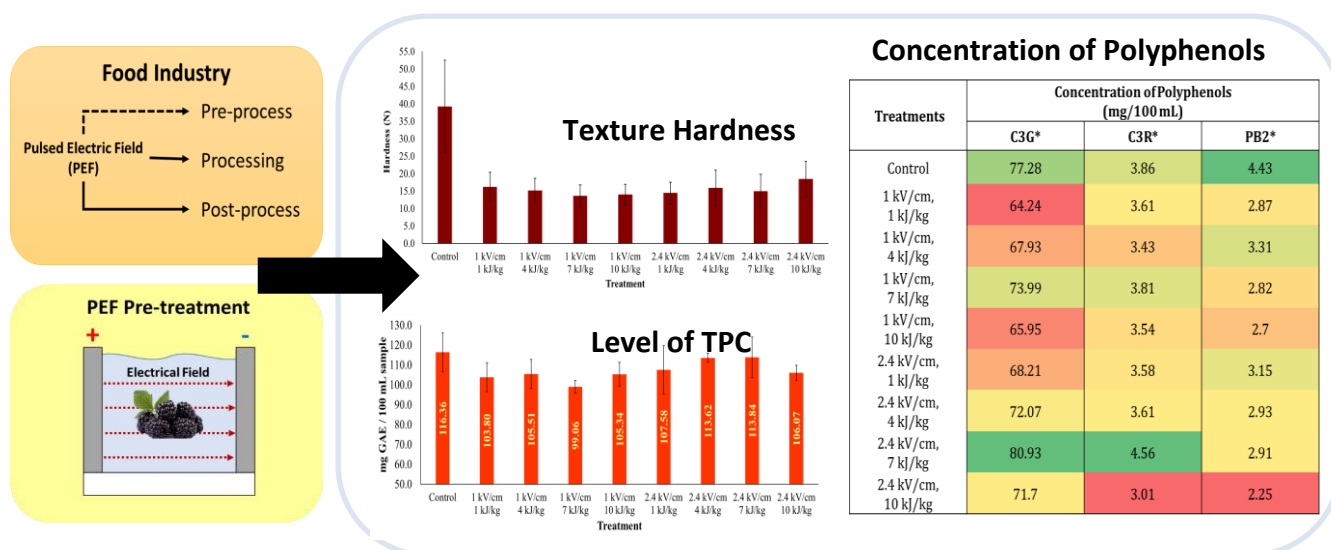
Abstract

The consumption of minimally processed Ready-To-Eat fresh produce has been recognised as a potential risk to human health. Fresh produce can become contaminated with foodborne pathogens *via* different transmission routes that could either be before harvest, i.e. pre-harvest sources (climate, geographical conditions, irrigation water, soil amendments, manure faeces and animal intrusion) or after harvest, i.e. post-harvest sources (processing practices, types of equipment, storage, transport, workers' hygiene, packaging and consumer handling). However, water used during the pre-harvest stage (irrigation water) and post-harvest stage (washing water) has been identified as one of the critical contamination sources of produce-associated foodborne illnesses. Furthermore, chlorination has been routinely used as a treatment method for disinfecting irrigation water and post-harvest cooling water. The objective of the present study is to create a Quantitative Microbial Risk Assessment (QMRA) to assess the human health safety risks (infection or illness) from the application of untreated irrigation water as opposed to that treated with chlorine as a water disinfection treatment and assess potential intake through the consumption of RTE fresh produce that is grown on the irrigated land. A quantitative model was constructed to analyse human exposure to foodborne enteric pathogens associated with the consumption of RTE produce. The model entails four steps- (i) Hazard Identification, where causative microorganisms capable of causing foodborne hazards in RTE produce were identified, (ii) Exposure Assessment, where a quantitative assessment of the likelihood of exposure to target pathogens was conducted through different model parameters, (iii) Hazard Characterisation, where a dose-response assessment was carried out using experimental data from foodborne outbreaks, and (iv) Risk Characterisation, where information obtained from exposure assessment and dose-response evaluation was integrated to estimate the distribution of risk. Under the conditions and assumptions of the study, the results revealed *Cryptosporidium parvum* as a contaminant of concern, followed by *Giardia lamblia* and Norovirus. There was a very low risk associated with bacteria based on conditions considered for the probabilistic modelling. This study may benefit farmers, consumers, and policymakers by prioritising the pathogens of concern to ensure food safety around RTE fresh produce.



Abstract

Pulsed electric field (PEF) is an emerging food technology that can be utilised in food processing. However, its application to pre-treat fruits is not widely explored. This study investigates the influence of PEF on the physical and chemical properties of pre-treated blackberries. Different levels of electric field strengths (or EFS; 1.0 and 2.4 kV/cm) and total specific energy inputs (or TSEI; 1, 4, 7 and 10 kJ/kg) were applied. Texture hardness, juice yield, total phenolic content (TPC), 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging and ferric reducing antioxidant power (FRAP) activities, as well as levels of predominant poly(phenols) were determined. PEF-treated blackberries had significant declines in hardness ($P < 0.05$), but no significant differences were found between PEF-treated samples. There was slight increases of juice yield in PEF-treated samples ($P \geq 0.05$). PEF treatment of 2.4 kV/cm EFS with 4 and 7 kJ/kg TSEI resulted in comparable levels of antioxidant indices (TPC, DPPH and FRAP) to untreated blackberries. The predominant polyphenols were cyanidin-3-O-glucoside (C3G), followed by cyanidin-3-O-rutinoside (C3R) and procyanidin dimer B₂ (PB2). The pre-treatment of EFS 2.4 kV/cm and TSEI 7 kJ/kg led to the highest content of C3G (80.93 ± 9.05 mg/100 mL) and C3R (4.56 ± 0.47 mg/100 mL) while the PB2 contents were low. With further optimisation, PEF pre-treatment can be applied to aid the blackberry juice processing.



Selected Recent Publications

- Putsakum, G., Tzima, K., Tiwari, B. K., O'donnell, C. P., Rai, D. K. (2023) 'Effects of thermosonication on ascorbic acid, polyphenols and antioxidant activity in blackberry juice', *International Journal of Food Science & Technology*, 58, 2304-2311.
- Tzima, K., Putsakum, G. & Rai, D. K. (2023) 'Antioxidant Guided Fractionation of Blackberry Polyphenols Show Synergistic Role of Catechins and Ellagitannins', *Molecules*, 17;28(4),1933.

COMPARISON OF PORTABLE INSTRUMENTATION FOR SPECTRAL IMAGING OF BACTERIA

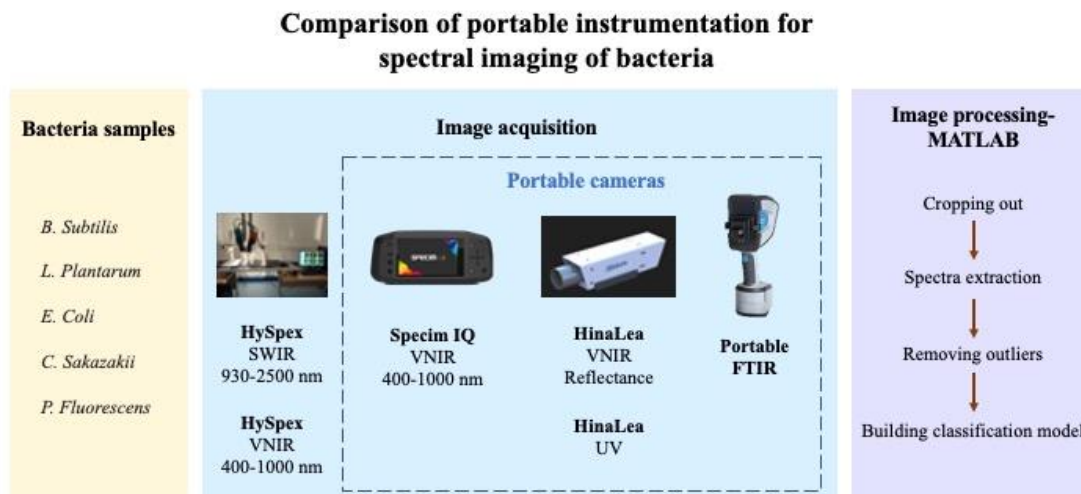
Fangting Bai, Aoife Gowen

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

Spectral imaging has broad applications in the food science field as the fast detection technology to control food quality, like the detection of bacteria. The traditional lab-based spectral imaging system has high image resolution; however, it is relatively expensive and bulky. Portable spectral imaging cameras could cover this shortcoming to realise on-site food quality assessment, which is online, rapid, and non-invasive. The main objective of this study is to compare the performance of the portable spectral imaging techniques applied to bacteria detection. In this study, the performance of the following portable systems has been compared: portable Fourier Transform Infrared (FTIR) spectrometer; 2 Visible and Near-Infrared (VNIR) spectral imaging cameras with different illumination setups, and traditional Short-Wave Infrared VNIR-SWIR spectral imaging system for comparison. Images and spectra were processed by chemometric methods (Linear Discriminant Analysis (LDA) and Partial Least Discriminant Analysis (PLS_DA)). Correct classification value and the pixel classification maps were the outputs of the models, which were used to judge the performance of the cameras. This study gave scientific support for the application of the portable spectral camera on bacteria detection.

Graphical Abstract



Introduction

Bacteria play an important role in the food industry. For example, they serve to ferment cheese and yogurt, but some bacteria will cause food spoilage and food poisoning as well. The identification of food-related bacteria is critical for food quality and safety. It is equally essential to detect pathogens and non-pathogens in the food production system (Wenning *et al* 2014).

Spectral imaging technology is the combination of spectroscopy and imaging technology, such that the spectral imaging camera can obtain image information and spectral information at the same time, with the advantages of being non-invasive, rapid, and informative. Nowadays,

spectral image technology has been broadly applied to bacteria identification and detection. Fourier Transform Infrared (FTIR) and Raman spectroscopy have been applied to obtain the spectra information of the *C. sakazakii* biofilm grown cells and the planktonic cells about the whole bacterial cell composition, the two techniques are relatively fast and less expensive than traditional spectra imaging system (Sharma and Prakash 2021). Traditional spectral imaging systems have high spectral and spatial resolution; however, they are relatively expensive and bulky, require high precision scanning opto-mechanical elements and computer control, and is inconvenient to carry (Yao *et al* 2019). Portable spectral imaging cameras could cover this shortcoming to realise on-site food quality assessment, which is online, rapid, and non-invasive as well.

The objective of the study was to compare the performance of the portable spectroscopic and spectral imaging techniques applied to bacteria detection.

Materials and methods

Sample preparation

Bacillus subtilis (*B. subtilis*) DSM 10, *Escherichia coli* (*E. coli*) DSM 11250, *Cronobacter sakazakii* (*C. sakazakii*) ATCC 29544, *Lactobacillus plantarum* (*L. plantarum*) DSM 20174, and *Pseudomonas fluorescens* (*P. fluorescens*). The dry cell samples on the stainless steel were performed according to one previous study (Xu *et al* 2022).

Table 1. Details regarding sample replicate 1 deposited on stainless steel.

Bacterial species	Slide size	The number of slides	The number of drops
<i>Bacillus subtilis</i> (BS)	L/S	4	16
<i>Escherichia coli</i> (EC)	L/S	3	14
<i>Cronobacter sakazakii</i> (CS)	L/S	4	16
<i>Lactobacillus plantarum</i> (LB)	L/S	6	24
<i>Pseudomonas fluorescens</i> (PF)	L/S	4	16

Image acquisition

Samples of bacteria were acquired by HySpex SWIR-384 spectral camera (NEO Ltd., Oslo, Norway) and the illumination is a customised DC linear light source. For the portable cameras, Specim IQ (SPECTRAL IMAGING LTD., Oulu, Finland) was used for image acquisition within Visible and Near-Infrared (VNIR) 400-1000 nm range with LED ring light, and HinaLea (4200C, HinaLea Imaging, USA) within VNIR 400-1000 nm range with LED ring light. FTIR spectra were obtained by Handheld FTIR Spectrometer (Agilent 4300 Handheld FTIR Spectrometer, Ireland).

Data processing

K-means method was used for the clustering bacteria and background as two clusters. Cluster I was the bacteria spectra. Cluster II was the background spectra. Then the bacteria pixel spectra and background spectra would be used to calibrate and test the Linear Discriminant Analysis (LDA) model. For comparison, the mean spectra of bacteria and background would be used for calibration and test the LDA model as well. The Partial Least Discriminant Analysis (PLS_DA) model would be trained and test for the prediction of the bacteria. The correct classification value (CC) was utilised to judge the performance of the model.

Results and discussion- *B. Subtilis*

Portable FTIR spectra and Principal Component Analysis (PCA) analysis

The FTIR spectra of *B. subtilis* and stainless stain were shown in Figure 1. Due to the large difference of the *B. subtilis* and the background, the spectra of FTIR could distinguish bacteria

from the stainless steel by the peaks at some specific wavenumbers. For *B. subtilis* FTIR spectra, there was a main broad band with a peak at 3300 cm^{-1} which corresponds to O-H and N-H stretching vibrations in protein and polysaccharide of the cell membrane, where the stainless steel had no specific peaks as expected. Assignment of absorbance bands for dry bacterial cells could be referenced from this article (Xu *et al* 2021). According to the Figure 1, the *B. subtilis* was distinguished by PCA as one cluster and the background was another cluster.

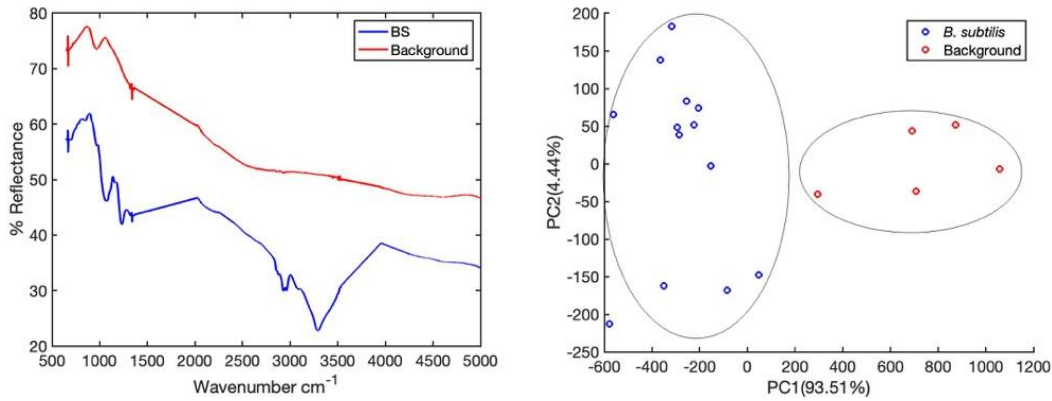


Figure 1. The direct reflectance of the mean FTIR spectra of *B. subtilis* on the stainless steel and the stainless-steel background (left). PCA score plot of FTIR spectra of *B. subtilis* and the stainless-steel background (right).

Table 2. The correct classification value of the LDA model of different instruments.

Rep 1	Hypex_ SWIR	Hypex_ VNIR	Hypex_ VNIR	SPECIM	HinaLea_ VNIR
Wavelength range	930-2400 nm	400-1000 nm	400-700 nm	400-700nm	400-700 nm
BS pixel spectra	72.83	88.01	84.79	91.97	98.54
BS mean spectra	93.75	100.00	92.86	93.75	93.75
EC pixel spectra	81.29	90.07	85.68	91.30	94.85
EC mean spectra	87.50	100.00	87.50	75.00	87.50
CS pixel spectra	81.96	92.13	88.69	88.19	87.24
CS mean spectra	85.71	85.71	50.00	81.25	75.00
LB pixel spectra	77.68	87.60	88.25	94.02	96.61
LB mean spectra	83.33	79.16	100.00	86.36	86.36
PF pixel spectra	89.39	89.84	87.10	84.43	95.39
PF mean spectra	100.00	43.75	75.00	87.50	100.00

LDA model-Correct classification value

Table 2 showed the correct classification value of LDA model on different bacteria using 5 different spectral imaging modalities. When pixel spectra were used to train the LDA model, usually they would get lower CC value than the mean spectra because mean spectra could represent the bacteria spectra and background spectra with less interference (however this was not always the case, for example pixel level models had higher classification accuracies than mean spectra models for HinaLea). The SWIR range was generally poorer than the VNIR for bacteria detection, while within the 400-700 nm range, the HinaLea system resulted in the highest accuracy for bacteria detection, except for CS whose highest CC value was obtained by Hypex VNIR.

Conclusions

Spectral imaging was successfully used for the bacteria detection. Portable spectral imaging cameras in the wavelength range 400-700 nm showed even better performance than the lab-based system for classification using LDA. FTIR spectra and PCA results presented the clear clusters of *B. subtilis* distinguished with the cluster of background. The results indicated that

portable spectral imaging cameras, could be used for detection of bacterial dry cells.

Acknowledgment

The master thesis was supervised by Professor Aoife Gowen, including the experimental design and the guidance of instrumentation, data processing, and draft writing. Thanks for Sakshi Lamba for preparing dry cell samples. Fangting Bai was responsible for image acquisition, data processing, and manuscript writing. The authors declare no competing interests.

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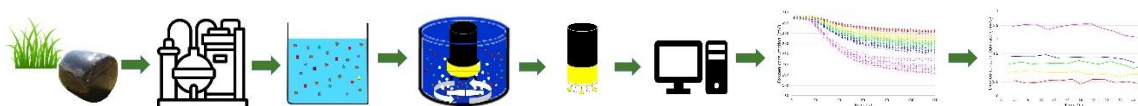
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Project Title: Development of a whole cell exclusion biosensor which is able to detect a whole range of analytes present in grass leachate.

Project Leader: Dr. Joseph Sweeney

Abstract

In Ireland there is an excess of 1.7 million tonnes of dry matter per annum of grass after the requirements for livestock production have been considered. The surplus of grass can even be increased by using marginal grasslands. Anaerobic Digestion is a technology that in recent years has been proposed as mechanism by which this surplus grass could be valorised to biogas, however, another grass valorising technology that has gained a lot of interest in recent years is green biorefinery (GBR). Grass silage fed GBRs operate by turning grass silage into an array of higher-value products, however, the quality of the silage dictates the quality of the products produced. While high-quality silages are rich in amino acids (AA) and lactic acid (LA) and result in higher-value products being produced, low-quality silages contain contaminants such as butyric- and acetic-acid and result in lower quality products being produced. Conventionally method such as high-performance liquid chromatography (HPLC), gas chromatography (GC), and near infrared (NIR) spectroscopy are used to measure key-performance analytes in bioprocesses and can be used to measure the quality of silage entering a GBR. However, a downside to technologies such as HPLC and GC is the fact that they are time consuming, expensive, and not automated. As most bioprocesses require that the concentrations of key-performance indicator analytes be measured as quickly and cheaply as possible, a real-time, cost-effective monitoring and control system is needed. This research aims to develop an array of *Escherichia coli* whole cell biosensors which will be used to detect key performance indicator analytes in real time, at-line within biological leachates produced from grass-silage fed GBRs. These biosensors consist of a bioreceptor which are *E. coli* cells that have been genetically engineered to “detect” a single key-performance indicator analyte with the exclusion of all other analytes present in solution. The exclusion of these interfering analytes is achieved by removing catabolic pathways within *E. coli*, that if present would result in *E. coli* catabolising them with oxygen as the terminal electron acceptor. The engineered *E. coli* is coupled to dissolved oxygen (DO) probe which is the transducer that turns the biological response into an electric signal. An *E. coli* biosensor presented with a specific analyte will catabolise said analyte. The oxygen depletion measured by the DO probe during this response will be mapped to analyte concentration, in solution. The fully developed biosensor system will allow for a robust and inexpensive at-line monitoring system which will enable the autonomously optimisation of the GBR process.

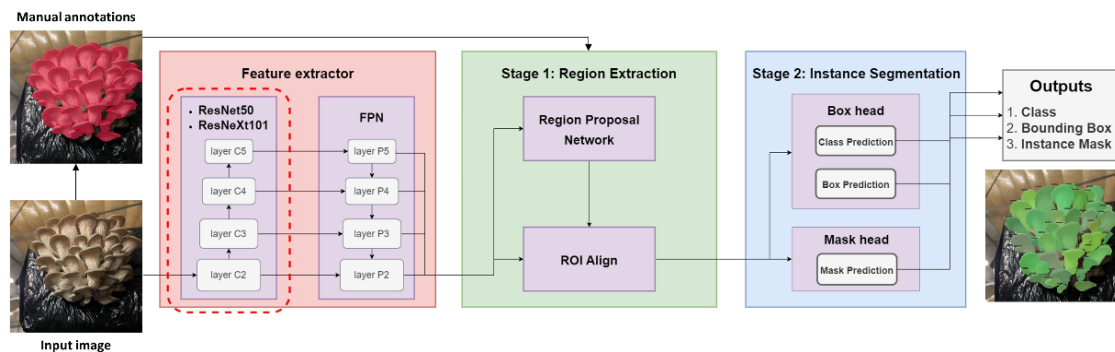


Project Title: Deep learning techniques for yield prediction in multi-domain mushroom production environments

Project Leader: Dr. Dimitrios Argyropoulos

Abstract

Accurate detection and localization of mushrooms under an actual production environment in order to enable automated crop growth monitoring still remains a challenge. Oyster mushrooms grow in clusters, which makes it more difficult to quantify their growth based on cap size and stipe length. The existing methods in practice mainly rely on weight measurement, which is time-consuming, destructive and labor-intensive. The aim of this study, therefore, is to design, test and validate a robust system to support detection and localization of mushrooms in a farm environment, helping farmers with yield estimation. Towards this objective, a comprehensive image dataset including different sizes, shapes and distribution densities was developed from Red-Green-Blue (RGB) images taken in an oyster mushroom farm. The proposed solution deals with the correct detection of mushroom clusters in an image while also precisely segmenting each mushroom using deep Convolutional Neural Networks (CNNs). Preliminary results showed that the Mask R-CNN based detection framework developed in this research can accurately identify and localize single oyster mushrooms in a cluster with adequate accuracy (mAP = 0.71). Future work will focus on field tests to monitor mushroom growth in an operational environment using time-lapse imaging techniques.



Selected Recent Publications

- Charisis C., Argyropoulos D. (2022) “A review on machine learning techniques in controlled environment food production systems,” In 36th EFFoST International Conference, November 7-9, Dublin, Ireland.
- Charisis C., Gyalai-Korpos M., Somosné Nagy A., Karantzaos K., Argyropoulos D. (2023) “Detecting and localizing mushroom clusters by a Mask R-CNN model in farm environment,” In 14th European Conference on Precision Agriculture, July 2-6, Bologna, Italy (in press).

DEVELOPING A POLICY LANDSCAPE TO SUPPORT THE DEVELOPMENT OF A GRASS TO PROTEIN BIOREFINERY CENTRE IN IRELAND

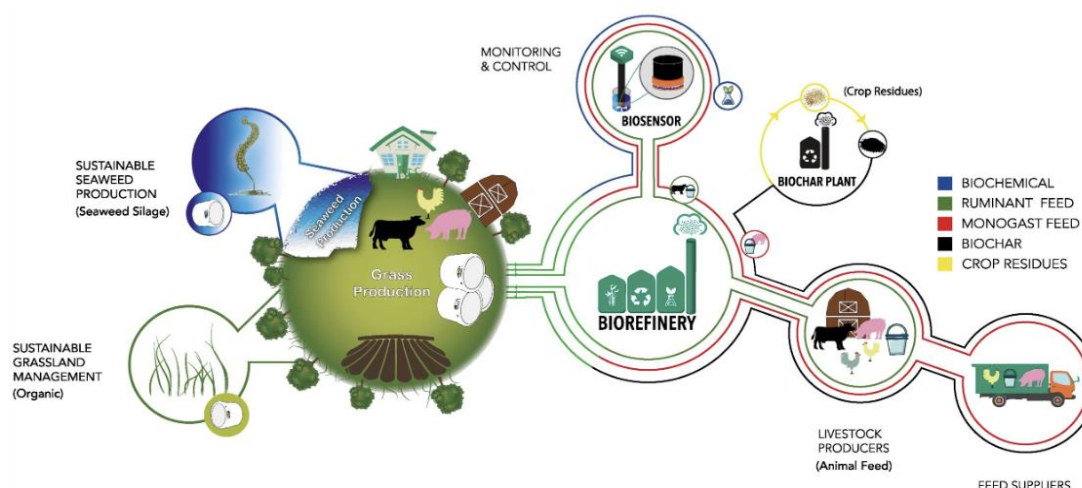
Lauren O’Riordan, Kevin McDonnell

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland

Abstract

The development and deployment of a novel protein production is needed in order to establish a sustainable food supply for both animals and humans. The EU’s food production system, from farm to fork, is responsible for 15% of net greenhouse gas emissions, however, 39% of food production related emissions which are estimated to occur outside of the EU through livestock feed imports are often unaccounted. Due to Ireland’s high proportion of livestock production to tillage area, the country relies heavily on livestock feed imports. While schemes such as the Protein Aid Scheme offers subsidies to farmers to grow crops rich in protein such as beans and peas, growing crops domestically will only produce a fraction of the protein needed for Irish livestock industries. The biorefinery system is a key strategy to address Ireland's protein demands and climate change crisis, however there is currently nothing around what legislation is needed for biorefinery on agricultural materials. The aim of the study is to develop a policy roadmap to support the development of a grass to protein biorefinery centre in Ireland. The study will examine and provide long-term policy instruments required to stimulate the growth of the biorefinery sector in Ireland. The study will draw heavily on the experiences of biorefinery development in other EU countries. The expected outcome of this project is to lay out a policy framework needed for biorefinery on agricultural materials which will provide economic and environmental advantages to Ireland’s agricultural and food sector.

Graphical Abstract



Biorefinery System Illustration (Farm4more 2020)

Introduction

Agricultural intensification has increased significantly in the last decade due to the pressure of having to feed another two billion people by 2043 (Ravindran *et al* 2021). Over 25% of global greenhouse gas (GHG) emissions are caused by the world’s food production system, from farm to fork, with food production in Europe alone being responsible for 15% of the continent's net GHG emissions (EEB 2019).

Livestock feed imports are a major share of agricultural emissions, but have remained hidden and unaccounted for in the EU, calling the overall sustainability of the sector into question (EEB 2017). In order to meet the growing demand for edible protein, the EU has become increasingly dependent on importing high protein livestock feed additives, such as soybean and maize, accounting for 39% of food production related emissions (EEB 2019). Sixty percent of Ireland's grain is imported for human consumption and animal feed (O'Brien 2023), however the war in Ukraine has caused a block on grain exports, leading the State to support farmers growing extra tillage and protein crops in order to make up for the shortage (Ravindran *et al* 2021). According to figures released by the Department of Agriculture, the Protein Aid scheme, which subsidises farmers to grow crops rich in protein such as beans and peas (DAFM 2022), has been shown to be the most cost-effective scheme for reducing carbon dioxide (CO₂) emissions from the agricultural sector (O'Brien 2023). However, even with the new schemes, arable crops are currently only meeting 35% of our national feed material needs (O'Brien 2023).

In order to achieve urgent climate targets, transitioning to low-carbon food production is of high importance for Ireland and the rest of the EU (Ravindran *et al* 2021). This has led to a huge focus on producing native protein which has been successfully demonstrated by the use of green biorefineries (Ravindran *et al* 2021). Ireland has more than enough raw material to supply a biorefinery system, as the country's total agricultural lands are 90% grassland, with the most commercially important grass being varieties of perennial ryegrass on which the ruminant sector is highly reliant on (Ravindran *et al* 2021). The green biorefinery process uses input substrates such as grass, legumes, silage and seaweed which are bio refined into a climate mitigating, economically viable, organic grass silage press cake used as animal feed (Farm4more 2020). Biorefineries also provide sustainable methods of generating multiple bioenergy products from various feedstock inputs, such as green chemicals used for fertiliser or for biomethane production in anaerobic digesters, therefore acting as a key mechanism to enable a circular economy (Ubando *et al* 2020).

Green biorefineries can enable Ireland to develop a native protein feed, reduce its dependence on imported feed materials and allow Ireland's food system to become more sustainable (Ravindran *et al* 2021). Green biorefineries are key to addressing environmental issues and with projects such as Farm4more, a grass-based bioeconomy in Ireland has the potential to be a real possibility in the near future (Farm4more 2020). Legislations such as the Protein Aid Scheme and Protein/Cereal Mix Crop scheme, the Common Agricultural Policy, Farm to Fork and UN sustainable development goals all focus on the development of sustainable food systems, however, there is currently nothing around what legislation is needed for biorefinery on agricultural materials in Ireland. This study will focus on providing a clear and significant insight into which stakeholders and policies are required to ensure the uptake and success of a green biorefinery sector in Ireland in order to produce a native protein animal feed and lower GHG emissions in the Irish food and agricultural sector.

The objective of the study is to develop a policy roadmap to support the development of a grass to protein biorefinery centre in Ireland.

Materials and Methods

The Life Farm4More agricultural pilot project, led by University College Dublin and co-funded by Life Programme and the Department of Environment, Climate and

Communications, is currently testing a press cake to feed dairy cattle in Ireland. The press cake is produced using silage grass in a biorefinery with the aim of reducing GHG emissions in the Irish agricultural sector (Farm4more 2020). This study will be working closely with the Farm4more project in order to develop a policy roadmap to produce a native protein animal feed in Ireland. A number of European countries are currently in the advanced stage of developing and implementing integrated biorefining pathways (IEA Bioenergy 2022). Table. 1 below highlights examples of biorefineries operating in Europe and current legislation introduced in order to move towards a circular economy while also strengthening economic development, supplying and safeguarding food quality, providing new income and reducing GHG emissions (IEA Bioenergy 2022).

Table 1. Biorefineries operating in the EU (IEA Bioenergy 2022)

Country	Facility	Input Substrate	Product	Legislation
Austria	AGRANA Biorefinery - Commercial	Wheat Maize	Animal feed Bioethanol	Austrian Bioeconomy Strategy (2019)
Denmark	GO-GRASS – Demo site	Grass	Animal feed	No national biorefinery strategy but established the National Bioeconomy Panel (2013)
Germany	BIOWERT - Commercial	Grass	Material Fertiliser	The New German Bioeconomy Strategy (2020)
Netherlands	Alco Energy Rotterdam Biorefinery - Commercial	Cereals	Animal feed Ethanol Electricity	The Biomass 2030 (2016) The Dutch National Climate Agreement (2019)

In order to successfully develop a policy roadmap to produce native protein animal feed in Ireland, the study will:

- Analyse data from current and various sustainable agriculture and food production legislation that would support and complement green biorefinery systems in Ireland.
- Analyse data from current legislation of EU countries who currently have pilot and commercially operating biorefinery systems.
- Analyse and identify carbon benefits from current policies and assess how grass biorefinery models can decrease agriculture and food system GHG emissions.
- Identify missing legislation and possible policy instruments within Ireland.
- Identify public and private stakeholders.
- Identify required collaborations across and within public and private sectors and their supply chains.
- Identify compliance benefits.

Results and Discussion

The study is in the early stages of development and the methodology of the study is open to modifications. As there is currently nothing around what legislation is needed for biorefinery on agricultural materials in Ireland, the expected results of the project are to provide a clear insight into which stakeholders and policies are needed and to provide linkages between existing policies that will help support the development of a grass to protein biorefinery centre in Ireland. The study will also discuss how a biorefinery model could enable Ireland to produce a native protein feed which will help lower GHG emissions in the agricultural and food sector and provide extra

income for farmers. Creating connections and cooperation is intrinsic to a successful circular economy and bioeconomy (Ubando *et al* 2020) and requires cross-sector and supply chain collaborations, which will be looked at in depth in order to successfully launch a biorefinery system in Ireland. The study will also discuss the necessary compliance benefits in order to ensure a successful biorefinery scheme uptake. A biorefinery system would ensure quality assured Irish meat/milk with lower carbon footprint which could possibly ‘piggyback’ onto Bord Bia’s Quality Assurance Scheme (Bord Bia 2023). The project will also look into the discrepancy of imported versus home grown local feed not taken into account by Bord Bia and how it could be considered. Potential consequences of increasing the area granted to protein crops under the Protein Aid Scheme (DAFM 2022) on the environment and biodiversity will be discussed, as well as the need for Ireland to use its importing market powers to insist the countries selling livestock feed are raising their environmental protection standards. The integration of effective policy, food and agricultural production agencies and conservation coupled with the application of scientific research should enable an optimal biorefinery system in Ireland and reduce GHG emissions.

Conclusion

Biorefineries can contribute to a more sustainable resource supply and help reduce Ireland’s overall GHG emissions. The development of long-term sustainable food and agricultural systems will rely on the monitoring and implementation of new schemes and strategies. By developing a policy roadmap to support the development of a grass to protein biorefinery centre in Ireland, the study will provide a clear and significant insight into which stakeholders and policies are required to ensure the uptake and success of the scheme and help reduce the hidden impacts that livestock import feed is having on our planet.

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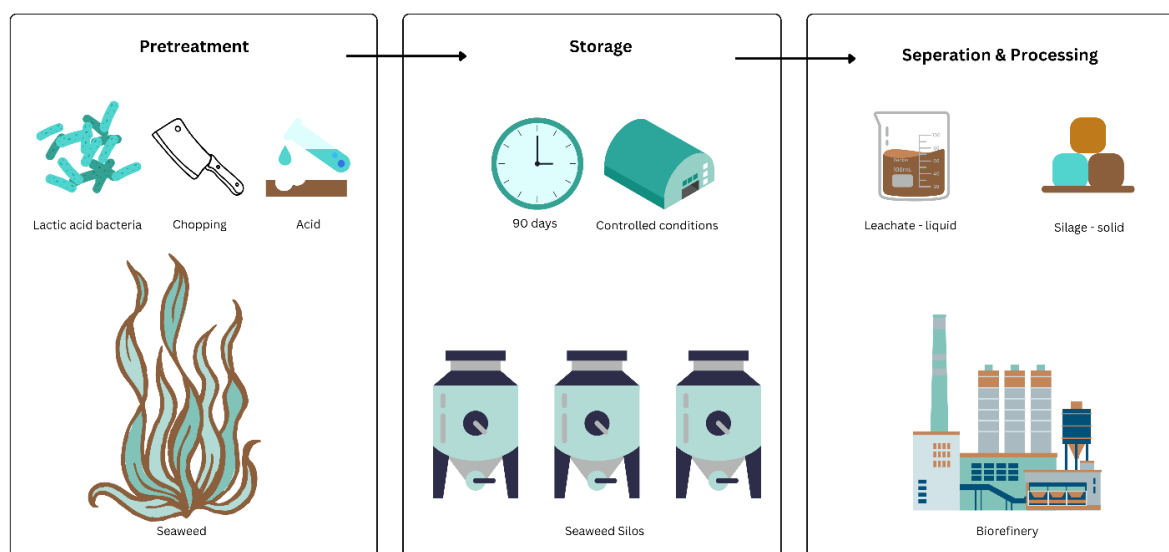
Priya D. Pollard, B.Sc., M.Sc.

Project Title: Ensiled seaweed as an alternative biorefinery input substrate

Project Leader: Dr Joseph Sweeney & Dr. Julie Maguire

Abstract

Blue farming initiatives are increasing Irish kelp production to better utilize coastal regions. Traditionally, harvested seaweed biomass is dried in ovens which contributes significantly to the total GHG emissions in the farming process. One way in which total farming emissions could be reduced is by preserving seaweed via ensiling as opposed to drying. This study aims to ensile seaweed to maintain the nutritional and monetary value of the biomass while producing a solid silage and liquid leachate both of which can be further valorized by biorefinery technologies. In this study large scale ensiling of *Alaria esculenta* and *Laminaria digitata* was conducted. Both conventionally produced kelps have high nutritional and monetary value and are currently used as sources of antioxidants, anticancer, anti-inflammation, antitumoral, bioactives, laxative and cardioprotective agents. In this study, seaweed biomass was optimally ensiled using a combination of *Lactobacillus plantarum* and citric acid. The homogenized mixture was compressed and stored for 90 days to determine ensiling stability. The seaweed-silage and -leachate quality was determined by measuring lactic and acetic acid levels. Additional analysis is necessary to determine the effects of the ensiling process on nutrient content, antioxidant levels, algin quantities and bacterial profiles. It is possible to ensile *A. esculenta* and *L. digitata* but more analysis is needed to valorise the products.



Selected Recent Publications

Pollard, P., Maguire, J., Sweeney, J., (2022) “Ensiling kelps for downstream biochemical extraction”, In Biorefine Conference ‘The role of biorefineries in European agriculture’, Belgium Ghent.

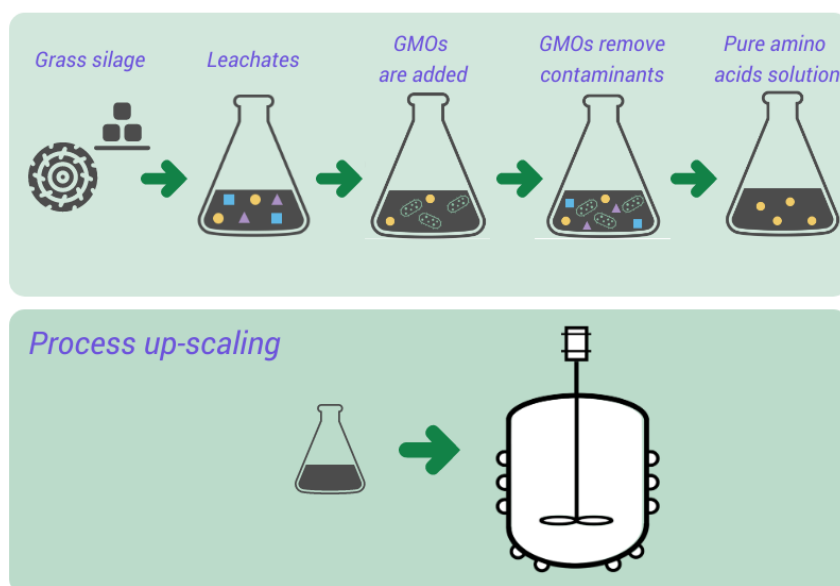
Anna Visentin, MSc

Project Title: Development of contaminant removal strains for the downstream purification processes in a green biorefinery

Project Leader: Dr. Joseph Sweeney

Abstract

There is an ever-increasing demand for amino acids (AA) for human food and animal feed consumptions, however, even though the manufacturing process has moved from the extraction of AA from natural sources to the chemical synthesis to biotransformation or fermentation processes (with 2 AA that are currently produced by chemical synthesis, 2 by extraction, 5 by biotransformation, 16 by fermentation), this still implies some drawbacks. Although fermentation and enzymatic routes have largely displaced the previous extraction methods, they generally use glucose, sucrose or molasses as carbon sources. The latter significantly impacts on the production costs therefore there is an increasing interest in evaluating alternative carbon sources. One of this could be grass, since grass silage-fed green biorefinery (GBR) leachates have been shown to be rich in AA. However, as grass silage leachates are the product of wild microbial fermentation, a number of impurities such as sugars, ethanol, butyric-, acetic-, propionic-, valeric-acid are also present in solution. To remove these contaminants and to produce purer AA products. A novel downstream purification process based on the use of genetically engineered *Escherichia coli* strains is proposed in this study. Gene deletions within a number of *E. coli* strains will be performed using the P1 phage mediated-gene knockout. Genes encoding for enzymes that need to be overexpressed will be cloned into high copy vectors and transformed into corresponding *E. coli* recipient strain. The strains obtained will be tested for their ability to grow aerobically on specific carbon sources and the performance verified by High Performance Liquid Chromatography (HPLC) analysis. The purification process will be upscaled from 250 mL Erlenmeyer flasks to a 2L bioreactor with the view of optimising the process. In the bioreactor parameters such as aeration and pH are controllable when compared to flasks and as such the performance of the contaminant removal strain is expected to be improved.



BIOBEO (FOOD LOOP, INNOVATIVE EDUCATION FOR THE BIOECONOMY)

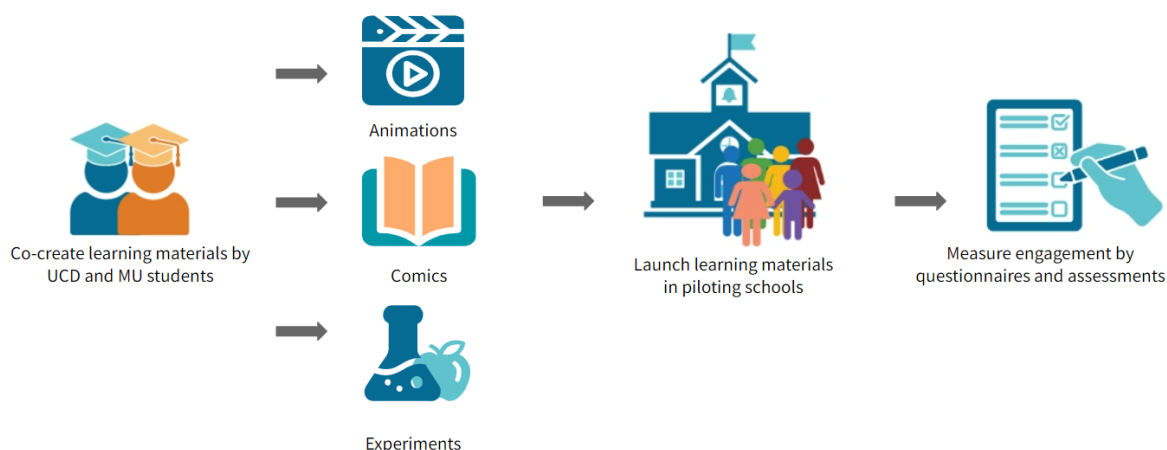
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Abstract

According to the Bioeconomy Strategy of the European Commission which aims to accelerate the deployment of a sustainable European bioeconomy to contribute towards Sustainable Development Goals (SDGs), the BioBeo, Innovative Education for the Bioeconomy, is established, to enhance understanding and engagement across society regarding circularity and the bioeconomy by introducing new approaches in education. The project is described through 5 themes which are Forestry, Life Below Water, Food Loop, Outdoor Learning and Interconnectedness. Education is the key factor to increase the engagement of young people and their intention to pursue education and careers in life science, technology and bioeconomy. This study aims to develop suitable learning materials, which are comics, animation and experiments about food loops for primary school students with measurable engagement from piloting schools. This paper provides the framework for developing learning materials in food loop themes under the BioBeo project based on the ADDIE development model. The acronym "ADDIE" stands for Analyze, Design, Develop, Implement, and Evaluate. Two main topics under this theme are food loss and food waste. Those materials are planned to launch in Green Schools in Ireland. The engagement and impact of this project will be measured by using questionnaires on target groups and assessment tests for students. The expected outcome of this project is to gain positive feedback from related participants, and measurable engagement to continually improve the learning materials. Further development is to provide gender and diversity-friendly manner materials.

Graphical Abstract



Introduction

The bioeconomy is a systems-based approach that seeks to replace fossil resources in a sustainable manner with renewable biological resources from terrestrial and marine ecosystems such as plants, animals and microorganisms to produce food, feed, fibres, energy, bio-based products, and services

within a circular economy framework designed to optimise resource usage based on a cascading hierarchy of utilisation possibilities (BioBeo 2022).

Currently, the world is facing climate change and other global challenges such as wars, food security, energy shortage and accessibility to education, all impacting sustainable development. To tackle these challenges, the application of education, scientific research, technology, and innovation with the aim of creating economic value, regenerating and expanding ecosystems and biodiversity are required. European Union decided to fund BioBeo project which aims to enhance understanding and engagement across society regarding circularity and bioeconomy by developing and deploying an education programme for children in different age groups from preschool to secondary school students. The aim of the project is to enable children to comprehend the resource base, including the growth timeline of specific plants at various stages that are suitable for product use. The fundamental concept behind this initiative is to instil the awareness that we, as human beings, "borrow" natural resources for the duration of our lifetime, and it is our responsibility to appreciate and preserve the environment. The project is described through five themes: forestry, life below water, food loop and outdoor learning and interconnectedness.

This paper will focus on the theme food loop in which learning material will be designed for primary school students. The term "food loop" refers to the process of producing, distributing, consuming, and disposing of food products. Therefore, this topic is relevant to various economic sectors such as agriculture, retail, and energy production. The key concept is to minimise waste and optimise the use of resources by keeping materials and products in use for as long as possible to ensure the sustainability of the food chain (BioBeo 2022).

The objective of this study is to develop suitable learning materials about food loops for primary school students with measurable engagement from piloting schools.

Materials and Methods

In the transition to a sustainable EU circular bioeconomy, education, together with research, knowledge and social participation, is a prerequisite (Golowko *et al* 2019). To instil awareness and understanding of sustainability effectively, the bioeconomy concept should be taught at a young age. The initial step towards achieving that is researching effective teaching techniques for children between the ages of 7 and 11. The combination of active learning and multisensory learning are effective teaching techniques for primary students (Bartholomew *et al* 2018). Comics, cartoons and experiments are chosen as learning materials for this project. In developing the aforementioned materials, the ADDIE model is adopted. The project will be co-created by students from University College Dublin and Maynooth University who study in sustainability and education respectively to develop suitable learning materials for children.

Analyse

This phase collects data on primary school students, aged 7-11, to understand their abilities and preferences. The food loop theme is introduced to help children grasp the resource base. Food loss and waste are chosen as subtopics as they relate to students' daily lives.

Design

The storylines, format and title are created regarding factors which are cultural barriers, such as diversity and language and educational network. The characters in the storyline should be inclusive to increase the engagement of students.

Develop

The editing process will use Adobe Flash. The content will be shared with teachers, parents, and students for feedback. Suggestions from media and educational experts will be sought. The learning materials will be revised based on the validator's feedback.

Implement

The finished product is planned to be launched in piloting primary schools that are organised by Maynooth University with effective teaching and learning methods from teachers.

Evaluate

Evaluation in piloting schools will be conducted through questionnaires and assessments. Questionnaires will gather feedback from students, teachers, and parents on enjoyment and effectiveness. Assessments, such as quizzes or tests, will measure students' understanding and retention of the learning material.

Food Loop

Globally, it is estimated that 14 percent of food is lost during the harvest process and 25 percent of food produced is wasted (FAO 2022). Food loss and waste (FLW) have adverse impacts on both food security and nutrition, and make significant contributions to greenhouse gas (GHG) emissions. EU countries are committed to meeting Sustainable Development Goal (SDG) Target 12.3, which aims to reduce global food waste at the retail and consumer levels and reduce food losses along production and supply chains, including post-harvest losses by 2030 (FAO 2022). Therefore, tackling this issue is a significant step towards a sustainable food loop.

The term "food loop" refers to the process of producing, distributing, consuming, and disposing of food products (OECD 2023). Therefore, this topic is relevant to various economic sectors such as agriculture, retail, and energy production. The concept of this theme is aimed to minimise waste and optimise the use of resources by keeping materials and products in use for as long as possible to ensure the sustainability of the food chain (BioBeo 2022). Figure 1 shows an example of learning material about food loss and waste for primary school students.

Food loss

Food loss occurs when the mass or quality of food decreases before reaching the consumer, often due to inefficiencies in the food supply chain. It can happen on the farm, in storage, during transportation. Examples include imperfect produce, surplus products, and expired food before sale (FAO 2022).

Food Waste

Food waste refers to food that was not ultimately consumed by humans and that is discarded or recycled (EPA 2018). Food waste can be the intentional or unintentional discarding of edible food which occurs at the retail and consumer stages. Examples of food waste include leftovers from a meal and scraps from food preparation such as bread crusts, potato skins, banana skins and chicken bones.



Figure 1. Example of learning material about food loss and waste for primary school students (Climate Science 2022).

Results and Discussion

This project is at the very early stages of development. The methodology and orientation are open to modification. There are 15 partners working on this project, the theme of food loops for primary schools will be co-created by University College Dublin and Maynooth University. Berlin Institute of Technology has specific expertise in delivering insight into the Food Loop theme as well. University College Dublin is mainly responsible for reviewing scientific literature, while Maynooth University is responsible for developing learning materials that are suitable for primary school students.

The expected results of this project are positive feedback from students, teachers, parents, and stakeholders, indicating increased engagement and understanding. The use of comics and animations is expected to enhance the connection between students and the bioeconomy. Food-related experiments aim to change values and raise environmental awareness. Measurable engagement from piloting schools will address any identified weaknesses. Increased societal involvement, from local communities to international levels, will drive progress towards a sustainable food loop.

In the future, after receiving feedback on the project, further developments which are gender and diversity-friendly manner materials and external events to present results and gather comments will be launched.

Conclusions

Effective teaching methods, coupled with suitable learning materials, enhance understanding and engagement in the bioeconomy. Comics, animations, and experiments will measurably impact primary school students' learning. The food loop theme aims to broaden children's perspectives on the value of food in the environment, economy, and society. Feedback from students, teachers, parents, and stakeholders will guide further development of gender and diversity-friendly materials for international use.

Acknowledgement

This research is being undertaken with the support of the School of Biosystems and Food Engineering at University College Dublin, Ireland.

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BIOBEO (INTERCONNECTEDNESS), INNOVATIVE EDUCATION FOR THE BIO-ECONOMY

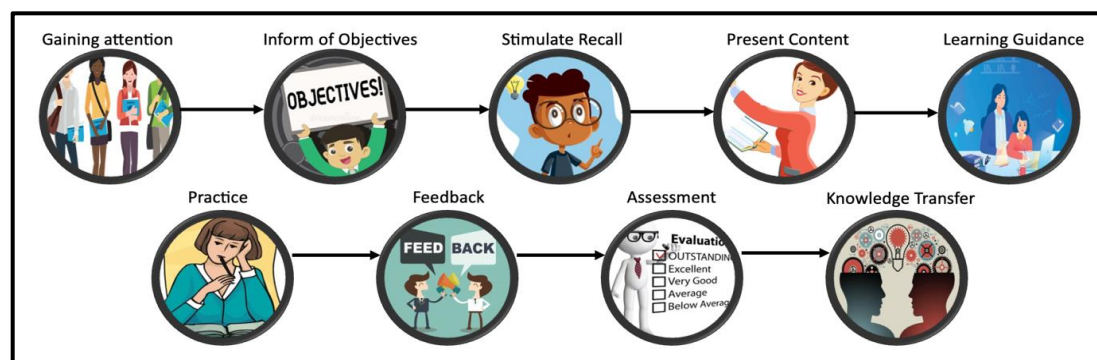
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Abstract

The BIOBEO project is an EU-based initiative that seeks to promote sustainable development through the bio-circular economy. The research explores how the integration of one theme; interconnectedness with the other 4 themes- forestry, life below water, food loop, and outdoor learning can create a symbiotic relationship to efficiently use resources, reduce waste, and protect the environment. One of its key themes is interconnectedness, which focuses on educating primary, secondary, and tertiary-level students about the interdependence of different sectors in achieving a bio-circular economy. This theme emphasizes the need for relevant policies and academic changes to instill a culture of sustainability among the secondary-level education generation. The concept of a bio-circular economy (BCE) is gaining traction as a promising approach for achieving sustainable development goals by introducing the effective utilization of renewable biological resources. BCE integrates biological and circular processes to create a closed-loop system where waste and by-products are transformed into valuable resources. The optimal scenario for educators would involve a solitary platform that seamlessly incorporates text, images, simulations, video, audio, and other multimedia resources into an integrated and cohesive setting that can be accessed from both school and home. This ideal can be achieved through the implementation of multimedia modules that are bolstered by Gagne's nine events of instruction. This method was chosen keeping in mind the concept of interconnectedness within the realm of the bio-circular economy is a relatively new concept and has potential yet to be discovered.

Graphical Abstract



Introduction

The Bio-Circular Economy (BCE) (Figure 2), unlike a linear economy (Figure 1) is a regenerative economic model that aims to create a closed-loop system where waste from one process becomes the raw material for another process. It is based on the principles of interconnectedness, which involves looking at the entire system from production and consumption to waste management and recycling. In this report, we will explore the theme of interconnectedness in the bio-circular economy, using the four other themes of forestry, life below water, food loop, and outdoor learning. Forestry, food loop, life below water, and outdoor learning are all interconnected and crucial components of a sustainable and regenerative system. The interconnectedness of the aforementioned are essential for the sustainability of our planet. Each of these elements plays a vital role in our environment, and together they form a complex web of interdependent systems. The thesis argues that the interconnectedness of BCE is crucial

for achieving sustainability via circularity. BCE creates a symbiotic relationship between different processes and stakeholders, allowing smooth functioning of the society and environment. The integration of biological processes, such as biotechnology, bio-resources, and microbiology, can enhance circular processes by converting waste into bio-based products and energy. It also promotes social equity by creating job opportunities and improving the livelihoods of local communities (Solanki *et al* 2014).



Figure 1. Linear – Economy.



Figure 2. Circular – Economy.

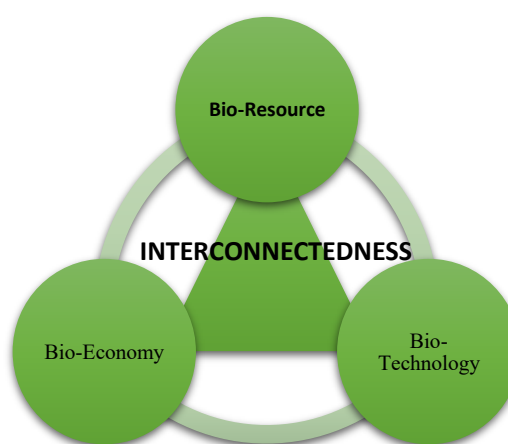


Figure 3. Interconnectedness bio-aspects.

In this report, we will explore the interconnected relationships between these four themes and their importance in promoting a more sustainable future.

Materials and Methods

BioBeo aims to establish crucial links between young people and their biological environment, as well as to study indigenous populations and their relationship with nature. This is vital for the future development of the bio-economy and circular processes (Figure 3), as industrial exploitation of the biosphere is a major contributor to climate change, which in turn reflects a lack of emotional connection to the natural world. BioBeo will focus on the biophilia hypothesis, which can potentially transform the mindsets and behaviour of European citizens [intervention at the earliest age possible for change], leading to measurable increases in well-being and holistic health. The consortium will develop innovative educational tools to support high-quality teaching, learning, and assessment, with a focus on raising awareness of the interconnectedness of all life on earth. These tools will require physical engagement with the earth, such as creating an indoor wormery, creating a comic book on the interconnectedness in BCE, and using various Science, Technology, Engineering and Math (STEM) learning tools, including SMART lab, augmented reality (AR) tv, Minecraft, and massive open online course (MOOCs) for Virtual reality (VR) training.

The objective of this research is also to substitute the epistemological idea of "waste" with a critical emphasis on refurbishment, reuse, and the decrease in the use of items that cause harm to the biosphere (Chodkowska-Miszczuk *et al* 2021). Nonetheless, the integration of the bioeconomy and circularity presents difficulties regarding its essence. This procedure involves the conversion of multiple sectors, the creation of novel value chains, the requirement to modify business models, and the encouragement of partnerships among ecosystems to achieve circularity (Salvador *et al* 2021). Hence, the creation of a BCE for the Urban Economy (UE) is contingent on determining its ability to fulfil requirements. If the UE were powered by renewable sources in an ideal scenario, discussions concerning the relevance of vital natural resources and energy efficiency would be less important. Nonetheless, ecological concerns and their urgent implications aside, urban planning and decision-making still underappreciated and neglect natural resources (Paes *et al* 2022).

A Look into Interconnectedness

Interconnectedness is the concept that all things in the world are interconnected (Figure 4) and have an impact on one another. *Forests* are a vital part of the ecosystem, and their health is directly linked to the health of life below water, helping regulate water cycles, maintaining soil fertility, and providing habitats for numerous aquatic species. When forests are destroyed or degraded, the health of rivers, lakes, and oceans is also affected. Similarly, the health of forests is also influenced by the health of *life below water*. For example, freshwater mussels help to filter water and improve its quality, which in turn benefits the trees and plants that grow on the forest floor. *Outdoor activities*, such as hiking, camping, and birdwatching, also rely on the health of forests and life below water. Without healthy ecosystems, these activities become less enjoyable and even dangerous. For example, hiking trails become muddy and slippery when there is too much runoff from degraded forests, while fishing and boating become less productive when the health of waterways is compromised. The *food loop* is another area where interconnectedness is important. The food we eat is grown in soil that is affected by the health of forests and life below water. If the soil is degraded or contaminated, its food will also be compromised. Similarly, how we produce and consume food impacts the health of forests and life below water. For example, industrial agriculture practices can lead to deforestation, water pollution, and soil degradation.

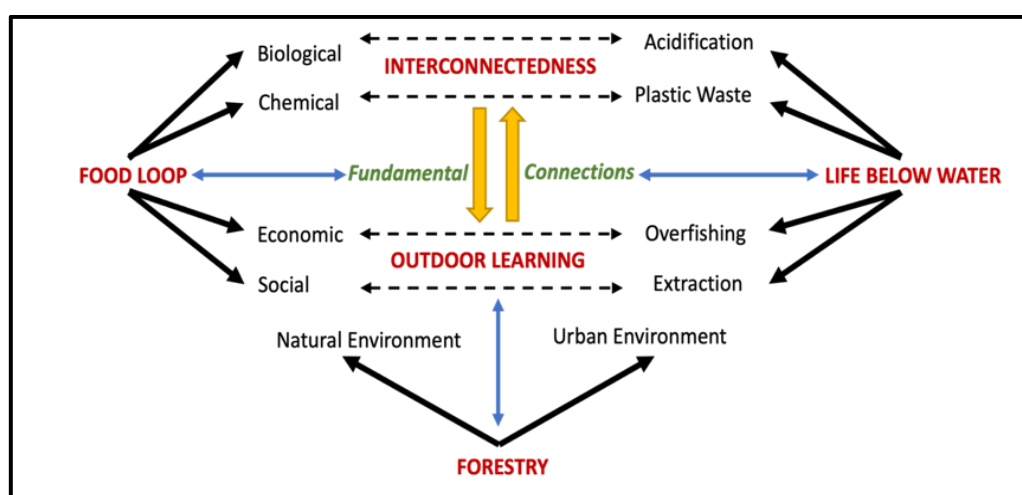


Figure 4. Interconnectedness among the five themes

Biophilia Hypothesis

Wilson (1984), Kellert (1996), and other researchers have proposed a daring theory, which is gaining more and more backing. This theory suggests that there is a fundamental and biologically driven human desire and inclination to connect with other living beings, known as biophilia. Children face various types of challenges in their daily lives, such as logical, mathematical, physical, social, ethical, and environmental problems. These issues necessitate

finding solutions to restore balance, as being in a state of disequilibrium is uncomfortable. Consequently, children aim to develop more effective ways of addressing problems, integrating different ideas, and comprehending the world around them. Environmental reasoning can be divided into two broad categories: homocentric and biocentric. Homocentric reasoning prioritizes the protection of the environment based on how it benefits humans, such as personal interests, aesthetics, and physical well-being. On the other hand, biocentric reasoning emphasizes that the natural environment has intrinsic value beyond its usefulness to humans and deserves protection based on its own moral standing. (Kahn *et al* 1997)

Results and Discussion

This thesis explores the interconnectedness of BCE and how integrating biological and circular processes can contribute to sustainable development. It commences by explicating the concept of BCE, providing an outline of its guiding principles, advantages, and obstacles. It further scrutinizes the interconnectedness of BCE from diverse perspectives, such as the economy, environment, and society with present instances of application in various industries, for example, agriculture, industry, rural and urban settings. Interconnectedness provides an in-depth exploration, emphasizing the importance of education in promoting sustainable practices, underscoring the importance of relevant policies and academic changes in instilling a sustainability-focused culture among younger generations and building a sustainable future for all. It also examines the potential of outdoor learning to promote sustainable practices and raise awareness. The integration of these themes can create job opportunities and improve the livelihoods of local communities. With amalgamation of the themes, efficient usage of resources, reduced waste, and environment protection is possible.

Conclusion

In summary, acknowledging the interdependencies between forestry, life below water, outdoor activities, and food cycle is crucial for establishing sustainable practices that benefit the communities and environment reliant on it. The interconnectedness theme of the BioBee initiative thoroughly explores the potential of BCE and highlights the significance of educating secondary-level students to promote sustainable practices. By integrating the five themes we can optimize resource utilization, diminish waste, and safeguard the environment. Inclusive and gender-neutral education materials will be developed to promote fairness and equality. Attaining sustainability necessitates the active participation of the community, parents, and positive feedback from stakeholders, emphasizing the need for a collaborative approach. Implementing relevant policies and academic modifications will foster a culture of sustainability among young learners and pave the way for a sustainable future.

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FORESTRY AND OUTDOOR LEARNING IN EUROPE

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Abstract

Outdoor learning is a concept that promotes the idea that education can be received outside the boundary of the confined space and through the BioBeo project my team plans to highlight how this concept can be utilized to put forward some of the current issues relating to climate change and its impact on our natural ecosystem. To achieve this goal my team and I are focused on developing simple laboratory protocols that can be performed outdoors to evaluate and quantify climate change's effects on forests, soil, and water bodies. These experiments include natural microbial activation in soil, testing the pH of water bodies, and determining whether a crop or plant is healthy or sick, and all these experiments are focused on generating results that will allow secondary school students to visualize and understand the topics and generate data while being present in the heart of the moment. Experimental safeguards will be considered one of the priorities while developing the protocols so that the instructor can execute the experiments while maintaining the safety of the students. The findings of this paper are to be adopted by secondary schools across the EU.

Graphical Abstract



Introduction

Outdoor learning promotes the belief that education can be acquired outside of a traditional classroom setting and that nature is the best source of knowledge (Türkmen *et al* 2010). In addition, learning outside enhances the growth of one's focus, emotions, cognitive skills, and intellect (Wells 2000). Indian Nobel prize winner Rabindranath Tagore believed that education should not be limited to classrooms and textbooks but should extend to the world around us. In that sense, his idea of learning in an open space was revolutionary (Mandal 2013). He advocated for a more experiential approach to education, where students could

learn from their environment and interact with nature. He believed that the natural world was an ideal classroom and that students could learn a great deal by exploring it.

Students could learn about flora, fauna, and overall ecosystems in these environments and develop a greater understanding and appreciation for the natural world. Open spaces can foster creativity and imagination. Being in nature could stimulate the mind and inspire new ideas and ways of thinking (Yıldırım and Özyılmaz Akamca 2017).

In a recent study, 35 youngsters over the age of five, were randomly selected for the study by the Turkish Ministry of Education and received schooling outdoors for 10 weeks. They were required to complete simple chores like making a nature album in which they gathered natural objects like twigs, stones, and flowers and categorized them according to their abundance and other factors like whether the leaves are rough or not, which flowers had an odor, and which didn't and these objects were glued to a notebook to keep track of the things they collected to make an album (Yıldırım and Özyılmaz Akamca 2017). The review determined that the program's goal of developing the abilities was accomplished and that improvements were seen as a result.

This paper aims to discuss how simple experiments can be set up outdoors, to give secondary school students a more vivid outlook on climate change and how it affects the forestry and natural ecosystem through the concept of outdoor learning.

Materials and Methods

Experiment Design Bioventing

Bioremediation can be carried out by students at sites of poor soil quality by bioventing (Alori *et al* 2022). The process of bioventing can be carried out by digging a narrow column called an injection well into the soil and by delivering oxygen from any form of the cylinder into the soil column thus allowing air circulation inside the soil and the oxygen in the air will enhance the indigenous microbial activity of toxin breakdown (Alori *et al* 2022) Regular observation needs to be carried out in terms of crop growth in areas where bioremediation is carried out as compared to where it is not.

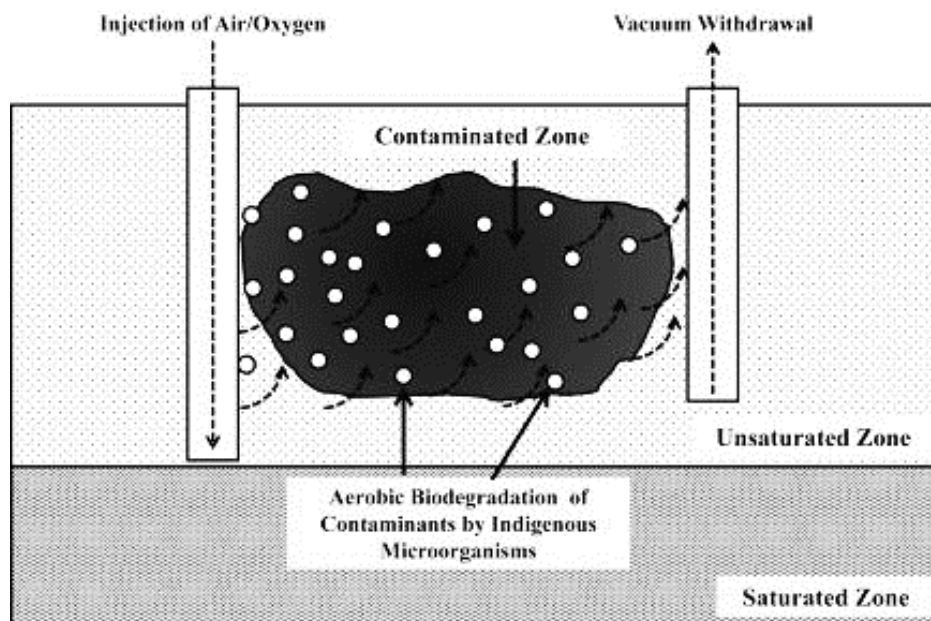


Figure 1. Bioventing schematic for microbial activation through oxygenation (Brown *et al* 2017)

By forcing air or oxygen flow into the unsaturated zone and, if necessary, by adding nutrients, bioventing increases the activity of native bacteria and archaea and stimulates the natural in situ biodegradation of hydrocarbon derivatives (such as, for example, forming a spill of crude oil or crude oil products) (Alori *et al* 2022).

Direct air injection may be used to infuse oxygen during bioventing into the soil with any remaining contaminants. As vapors move slowly through biologically active soil, bioventing primarily aids in the breakdown of adsorbed fuel residuals, but it also aids in the degradation of volatile organic compounds (VOCs) (Speight 2020).

Students need to carry out these procedures under the supervision of a teacher with sound knowledge and experience in equipment handling. Local support from farmers is required for regular reports on crop growth profile in the remediated area.

Checking the pH of the water

The pH of water for irrigation needs to be between 6 to 8 to support crop growth and freshwater pH needs to be between 6.5 to 9 to support aquatic life. Any deviation of pH beyond or below this range can cause serious damage to the aquatic ecosystem and farmland, including crops and soil.

To determine the pH of water students can conduct simple acid-base titration using color change as endpoints to determine if the water is severely acidic or alkaline. A simple plastic burette, pipette, and beaker can carry this experiment outdoors as harmless reagents like phenolphthalein and methyl orange. These reagents are sensitive to both strong and weak acid-base combinations and hence can detect the acidity or alkalinity of water from different sources.

Teachers need to make sure that the students are well informed of the titration principles and methodologies and how to record and interpret the endpoint of the reactions based on color change. Instructors also need to make sure that the experimental setup is established on a flat solid surface to prevent any sort of spillage prevent reagent wastage as the experiment is being conducted outside alab and it will be inconvenient in case there is a reagent shortage due to wastage.

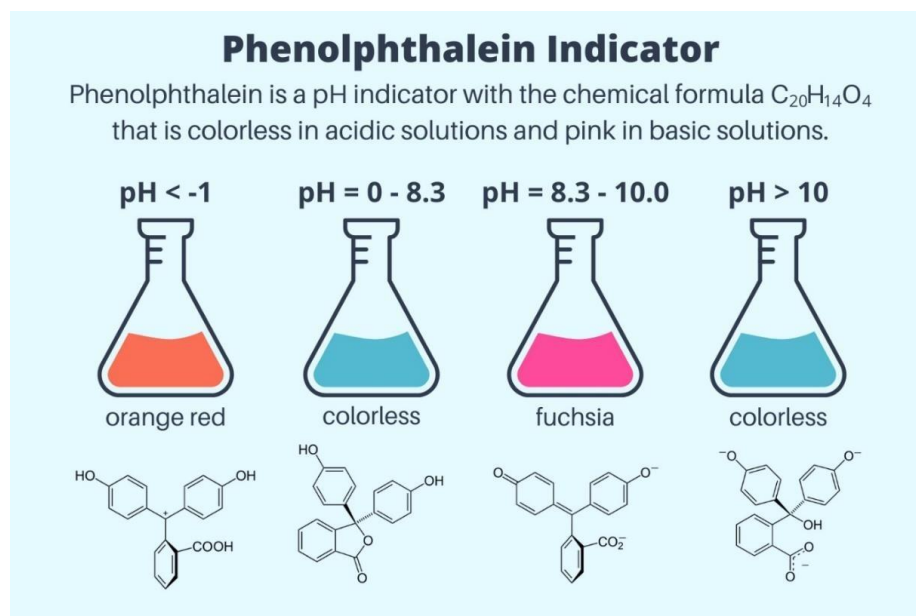


Figure 2. Endpoint titration colour change shows the acidic or basic nature of the water source (Helmenstine 2022)

Results and Discussion

These experiments aim to make students more aware and conscious of nature and understand how globalization, industrialization, and modernization in the 21st century affect nature. With the data and results obtained from these outdoor activities, students and teachers can seek help from local authorities by providing them with evidence of the damage being done, and through the involvement of a larger community, try to work out solutions for remediation.

Forestry and outdoor learning are the section of this project which will try to tackle some of the existing problems in nature by practicing science in nature to create a bigger impact through visual engagement and on-site data assessment and evaluation.

Conclusions

The results obtained from the designed experiments will be used to better clarify the effects of climate change and establish the need for sustainability by providing concrete evidence. Students will be encouraged to conduct their experiments in nature to highlight some of the issues that they think are evident in their local area and try to generate awareness throughout the community. Experimental data can be used to convince local authorities and businesses to reduce their emissions and enlighten people to adopt eco-friendly habits in their daily life.

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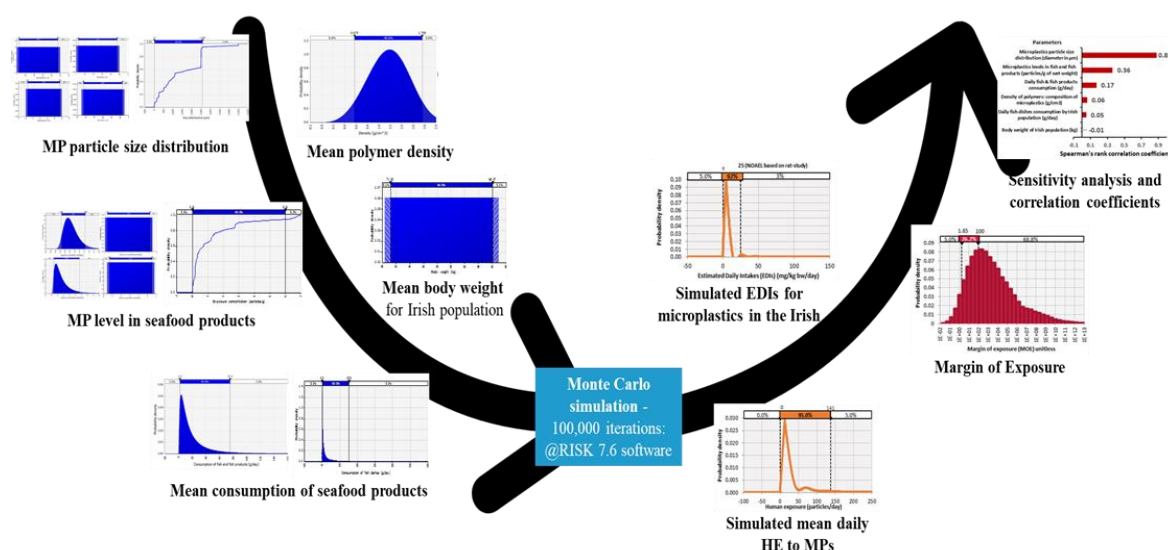
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Project Title: Human health risk assessment of microplastics through seafood products

Project Leader: Prof. Enda Cummins & Dr. Rajat Nag

Abstract

Microplastics (MPs) may pose toxicity issues, including acute and (sub) chronic toxicity, carcinogenicity, and developmental toxicity. Ingestion of MPs has been identified as the predominant exposure pathway. This study conducted a probabilistic exposure assessment on the potential human exposure (HE) to MPs through fish and fish products. These products are reported to result in a higher share of HE to MPs through the ingestion pathway. This research revealed that the simulated daily mean HE to MPs through fish and fish products consumption was 32 particles per day. Based on the Estimated Daily Intakes (EDIs) of the Irish population, the simulated mean EDIs to MPS was found to be 4 mg/kg bw/day. Results indicate there is a 97% probability that the HE to MPs will not go beyond the no-observed-adverse-effect level (NOAEL) value of 25 mg/kg bw/day. With the Margin of Exposure (MOE) approach, 68.8% of the population is at low risk to MPs exposure through fish and fish products. A sensitivity analysis revealed that the microplastics particle size is the most sensitive parameter of the model. This research will inform the public, food growers, and policymakers on potential exposure to MPs and risk reduction measures, thus, ensuring food safety and encouraging responsible use and disposal of plastic materials.



Selected Recent Publications

- Yuan, Z., Nag, R., Cummins, E. (2022) 'Ranking of potential hazards from microplastics polymers in the marine environment', *Journal of Hazardous Materials*, 429(128399), 1–19.
- Yuan, Z., Nag, R., Cummins, E. (2022) 'Human health concerns regarding microplastics in the aquatic environment - from marine to food systems', *Science of the Total Environment*, 823 (153730), 1–19.
- Yuan, Z., Nag, R., Chhaya, R.S., Cummins, E. (2022) 'Human health risk assessment of microplastics through seafood products', in *12th Biennial Conference (FOODSIM2022)*, Ghent, Belgium.

HUMAN EXPOSURE TO BPA ANALOGUES THROUGH FOOD

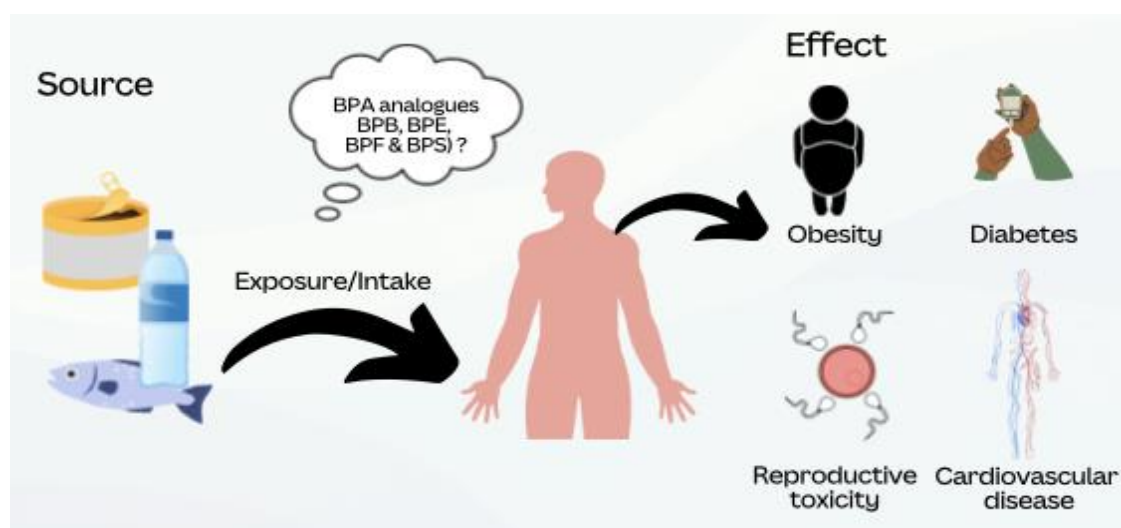
Erika Vázquez Vela, Xin Wang, Enda Cummins.

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

For this study, a risk assessment is performed using hazard identification, hazard characterization, exposure assessment and risk characterization. The human health risks from BPA analogues were identified, specifically for given populations and the main risk factors for contamination of food supply chain, in order to evaluate the likely level of human exposure to BPA analogues, while considering consumption patterns. This study will develop a model that will show that the human exposure to BPA analogues (specifically B, E, F and S) through food products is below any regulatory and toxicity level, posing a negligible risk to the consumer.

Graphical Abstract



Introduction

New risks arise from the expansion of food technology and production that need to be assessed and researched (Jenkins *et al* 2020). Worldwide, food products are produced and then distributed, guaranteeing food quality and safety through regulations and policies (Gallart-Ayala *et al* 2011). One of the most indispensable determinants of human health is food, which is the reason behind its increase in research.

Bisphenols include many substances that have a similar chemical structure, with a common two phenolic rings that are joined together through a carbon (Rivas *et al* 2002). There are at least twenty bisphenol analogues, such as bisphenol B (BPB), bisphenol E (BPE), bisphenol F (BPF), and bisphenol S (BPS (Zhao *et al* 2023).

Bisphenol A or BPA has one of the highest production in the world, with a growing demand over the past decades of millions of tons (Wang *et al* 2022). Though it has been associated with a wide range of adverse health effects, from reproductive disorders (fertility and development, prenatal or early postnatal exposure) (Milić *et al* 2015), obesity, especially in children and adolescents), and cardiovascular disorders (Kang *et al* 2006).

Manufacturers have been seeking BPA alternatives for “BPA-free” products (Wang *et al* 2022). In an attempt to stop the BPA use due to their health risks, BPA alternatives have been developed, which include bisphenol analogues (Zhao *et al* 2023). Some applications for

bisphenol analogues include plastics and resins (Gallart-Ayala *et al* 2013). For example, BPS has been used in food and beverage metallic cans (Ye *et al* 2015), mainly used for its inner surface (Eladak *et al* 2015). Humans can be exposed to BPA analogues, mainly through ingestion by the consumption of food and beverages, which are stored in plastic containers, bottles, and cans (Chen *et al* 2016).

The objective of this study is to determine the human exposure to BPA analogues (specifically B, E, F and S through food products is below any regulatory and toxicity level and poses negligible risk to the consumer, while developing a risk assessment model and using the principles of quantitative risk assessment and using Monte Carlo simulation model.

Materials and Methods

Risk analysis is used to manage food risks, composed of risk assessment, risk management and risk communication (Jenkins *et al* 2020). Risk analysis gathers information and evidence on the risk level of a certain contaminant in the food supply, while identifying control points along the food chain (WHO 2006). Risk assessment is the first step of risk analysis (WHO 2006), which is the science of understanding hazards, as well as the occurrence and the consequences (Jenkins *et al* 2020).

According to the Food and Agriculture Organization, risk as a function of the probability of an adverse health effect and the severity of that effect, consequential to a hazard in food (FAO 2019). Risk assessment has four analytical steps (WHO 2006): Hazard identification, Hazard characterization, Exposure assessment and Risk characterization.

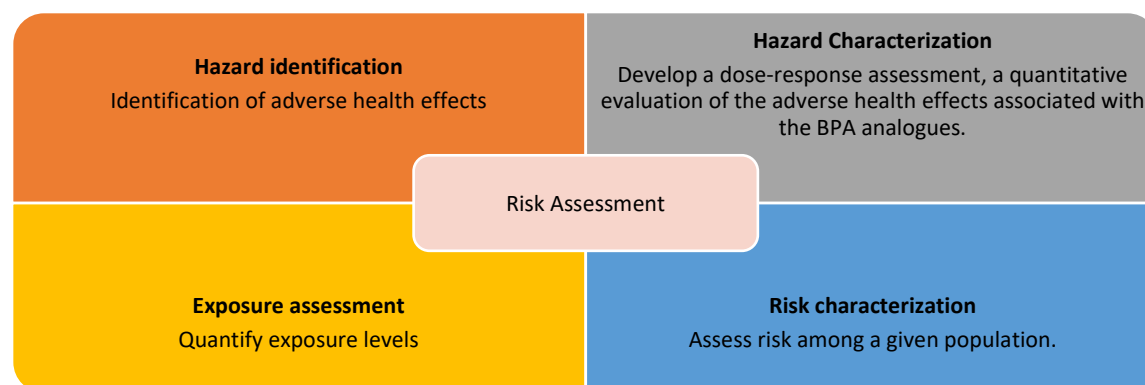


Figure 1. The framework of risk assessment (WHO 2006).

For this study, a risk assessment framework shown in Figure 1 is used, in order to develop a model to make sure that the human exposure to BPA analogues (specifically B, E, F and S) through food products is below any regulatory and toxicity level and poses negligible risk to the consumer.

The human health risks from BPA analogues were identified, specifically for given populations and also the main risk factors for contamination of food supply chain, in order to evaluate the likely level of human exposure to BPA analogues., while considering consumption patterns.

The information was collected from scholarly databases such as Google Scholar, Web of Science, and ResearchGate. The main search keywords used were ‘BPA analogues’ and ‘food products’, as well as ‘health effects’, ‘toxicity’, ‘adverse outcomes’, ‘disease’, ‘dose-response’ and ‘safe’. Guidelines and regulations were also identified, and literature contained keywords including ‘BPB’, ‘BPF’, ‘BPE’, ‘BPS’, ‘contamination sources’, ‘exposure routes’, ‘occurrence’, ‘packaging’, ‘resins’, ‘can’, were considered for investigating BPA analogues.

Results and Discussion

As BPA levels in human samples have declined, due to regulation, the use of BPA analogues, such as BPS, has increased in the same ones, thus replacement compounds should continue to be monitored closely (Jacobson *et al* 2019). Alternatives for BPA in industrial applications, mainly include BPS, and BPF with large-scale applications (Zhao *et al* 2023).

Human health risks were identified, a study conducted between 2000 and 2014 in the United States, showed that the exposure to BPS was increasing (Rancière *et al* 2019). According to (Eladak *et al* 2015), BPS and BPF show antiandrogenic effects, as a harmful effect in humans and rodents, that are similar to those of BPA. Many epidemiological studies report that there is an association between BPF and BPS exposure and diabetes (Oliviero *et al* 2022). Some studies indicate that BPA analogues can cause the same type of cardiovascular diseases as BPA (Abrantes-Soares *et al* 2022).

BPA analogues can affect populations in a different form, as well as consumption patterns differ. They could be identified as potential chemical obesogens, that enlarge adipocytes, and promote adipocyte differentiation, inducing obesogenic effects in humans (Jacobson *et al* 2019). BPF can be associated with obesity, especially in children and teenagers (ages 6-19 years) (Jacobson *et al* 2019). BPA analogues can affect fetal and maternal health, and there could even be transfer from the mother to the fetus via the placenta (Abrantes-Soares *et al* 2022).

At any point of the different stages of the supply chain during the processing, packaging, transportation and storage of food, chemical contamination can occur, but quantifying and at what point of the chain can be difficult (Choudhury *et al* 2022). Humans can be exposed to BPA analogues, mainly through ingestion by the consumption of food and beverages, which are stored in plastic containers, bottles, and cans (Chen *et al* 2016). BPF can be accumulated through the supply chain (Eladak *et al* 2015).

According to Chen *et al* (2016), dietary sources have been reported as the highest contributor to BPA analogues exposure, rather than non dietary sources, though BPA analogues are present in multiple routes. There is not necessarily regulation specifically for each of the bisphenols analogues, so other regulations will be used for the comparison between the quantitative data that was obtained in the model and the regulations. The EU Regulation No 10/2011 establishes that plastic materials and articles that are intended to come into contact with food should not transfer their constituents to food simulations in quantities that exceed 10 mg of total constituents released per dm² of food contact surface (mg/md²). Additionally, it is stated that for food intended for infants and young children, plastics materials and articles should not transfer their constituents to food simulants in quantities that exceed 60 mg of total constituents per kg of food simulant (mg/kg).

There are not specific toxicity levels, maximum permitted levels, maximum residual limits, tolerable daily intake and others, for all the BPA analogues, so the results obtained in the model may not be compared to all the data. The expected results from the quantitative modelling approach that used risk factors, consumption patterns and different groups, will show that the human exposure to BPA analogues (specifically B, E, F and S) through food products is below regulatory and toxicity limits, though they are a negligible risk to the consumer.

Conclusions

In an attempt to stop using BPA, manufacturers have been seeking alternatives, which include bisphenol analogues, such as BPB, BPE, BPF, and BPS. While there are several gaps in literature regarding the BPA analogues, the expected results from the quantitative modelling approach, will show with the given data, that the human exposure to BPA analogues (specifically B, E, F and S) through food products is below regulatory and toxicity level, though they are a negligible risk to the consumer.

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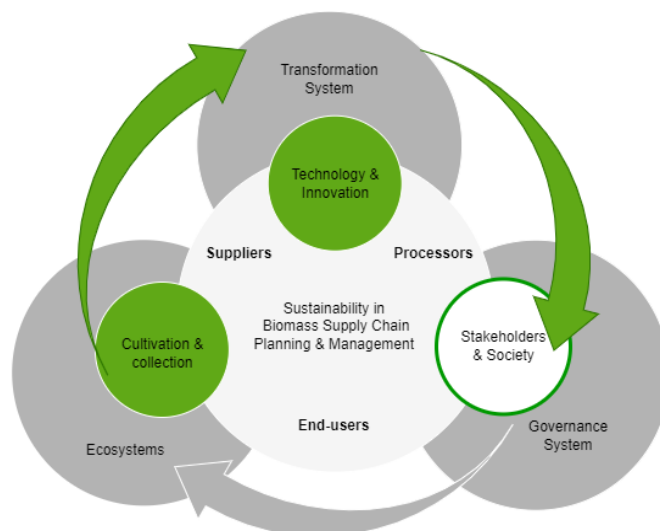
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Project Title: Strategies for biomass supply systems in the context of an emerging circular bioeconomy: Inclusion of human dimensions in sustainability transformations

Project Leader: Dr. Fionnuala Murphy

Abstract

In the European context, the sourcing of renewable bioresources and their conversion into value-added products such as food, feed, biochemicals, textiles, natural fertilizers, and bioenergy is being supported by several policies while seeking the development of a circular and sustainable bioeconomy. This will give rise to the expansion of new bio-based supply chains, which are closely related to the availability of bioresources and strategic planning. However, current approaches have been disregarding social aspects while largely focusing on technical-economic or environmental aspects at strategic decision-making levels. The objective of this research is, therefore, to address the social dimension of sustainability with forward-thinking perspectives for the planning process of new supply systems involving the sourcing of bioresources, illustrated by the potentials provided by grass and seaweed in biorefining. By adopting a mixed-method approach, both in-depth understanding, as well as broader representation, are possible with the combination of qualitative and quantitative analysis and methods. The results identify a broad range of aspects currently disregarded in the strategic planning of biorefineries, for example, farmer participation, conflicts over natural resources, collaborative capacity, quality or availability of jobs, gender equality, education and training. Those aspects raised the need for diversity in theoretical and methodological approaches as well as interdisciplinarity. In the case of new supply chains based on seaweed, upscaling cultivation would be required which entails numerous bottlenecks and social responsibilities of emerging enterprises over ecosystems, coastal communities, users of the marine space and consumer segments. The results could be used in the decision-making processes of practitioners, policy-makers, and grounds for future research in what relates to the planning and management of supply chains based on bioresources that ultimately are inclusive of human dimensions of sustainability.



Selected Recent Publications

- Cerca, M., Sosa, A., Murphy, F. (2023) 'Responsible supply systems for macroalgae: Upscaling seaweed cultivation in Ireland', *Aquaculture*, 563(P2), 738996.
- Cerca, M., Sosa, A., Gusciute, E., Murphy, F. (2022) 'Strategic planning of bio-based supply chains: Unlocking bottlenecks and incorporating social sustainability into biorefinery systems', *Sustainable Production and Consumption*, 34, 219–232.

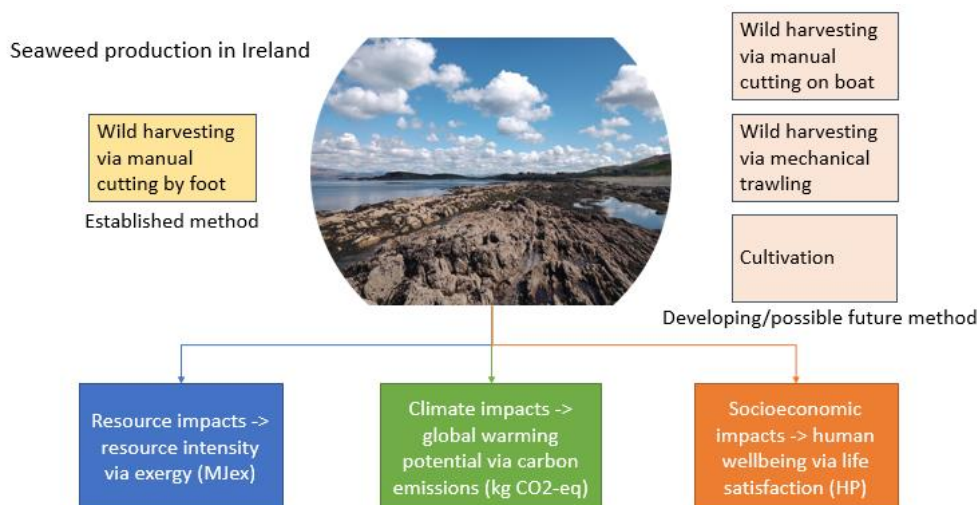
Charlene Vance, B.Sc., M.Sc.

Project Title: Understanding potential environmental and social impacts of Irish seaweed production through life cycle assessment

Supervisor: Dr. Fionnuala Murphy

Abstract

Macroalgae, or seaweed, is a versatile bioresource which can be used as a source of food, feed, fertiliser, and higher-value products. While production is currently done at smaller scales in Europe, demand for seaweed is expected to grow, and countries such as Ireland have the potential to produce significant volumes domestically. This study aims to capture the environmental and social consequences of different pathways for scaling up Irish seaweed production. Production pathways considered are wild hand harvesting by foot, wild hand harvesting by boat, wild mechanical harvesting, and cultivation. Several life cycle assessment (LCA) methodologies are utilised: exergetic LCA for impacts on resources, environmental LCA for climate change, and a novel social LCA framework for impacts on human wellbeing. The results demonstrate clear trade-offs in both environmental and social dimensions. Despite a large extraction of natural resources, wild hand harvesting was found to have a relatively low climate impact and resource intensity per kg seaweed produced. With mechanisation, climate impact and resource intensity increase significantly. For cultivation pathways, climate impact and resource intensity are highest for the small-size farm, decreasing as farm size increases. On the other hand, the small and medium-size farms are more labour intensive than large-size farms and thus provide the most social benefits per kg seaweed produced. Ultimately, when it comes to scaling up Irish seaweed production, pathways should be assessed based on the overall priorities of society.



Selected Recent Publications

Vance, C., Sweeney, J., Murphy, F. (2022) 'Space, time and sustainability: The status and future of life cycle assessment frameworks for novel biorefinery systems', *Renewable and Sustainable Energy Reviews*, 159, 112259.

Vance, C., Mainardis, M., Magnolo, F., Sweeney, J., Murphy, F. (2022) 'Modeling the effects of ecosystem changes on seagrass wrack valorization: Merging system dynamics with life cycle assessment', *Journal of Cleaner Production*, 370, 133454.

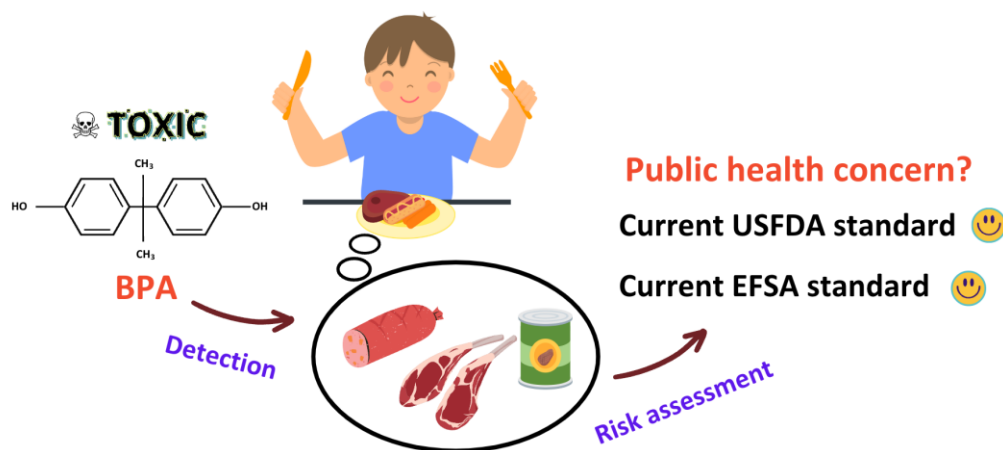
Project title: Comparative risk assessment study on bisphenol A (BPA) through meat products

Project leader: Professor Enda Cummins, Dr Rajat Nag

Abstract

Bisphenol A (BPA) is an endocrine disruptor associated with multiple adverse effects on human health. Dietary consumption is the predominant route of exposure considering BPA has been frequently detected in different types of food, including meat and meat products. The aim of this study is to assess the risks of BPA exposure from meat consumption. Probabilistic models were developed based on the literature-based BPA concentrations in canned and non-canned meat products and consumption rates of meat products among Irish consumers. The simulated mean (95th percentile) exposure levels were 3.5E-03 (1.4E-02) and 2.5E-04 (8.5E-04) $\mu\text{g (kg bw)}^{-1} \text{ day}^{-1}$ for non-canned and canned products among adult consumers, respectively. There was a low risk of BPA exposure from meat consumption according to the current health-based guidance values of 5 and 4 $\mu\text{g (kg bw)}^{-1} \text{ day}^{-1}$ set by U.S. Food and Drug Administration and European Food Safety Authority (EFSA), respectively.

Graphical Abstract



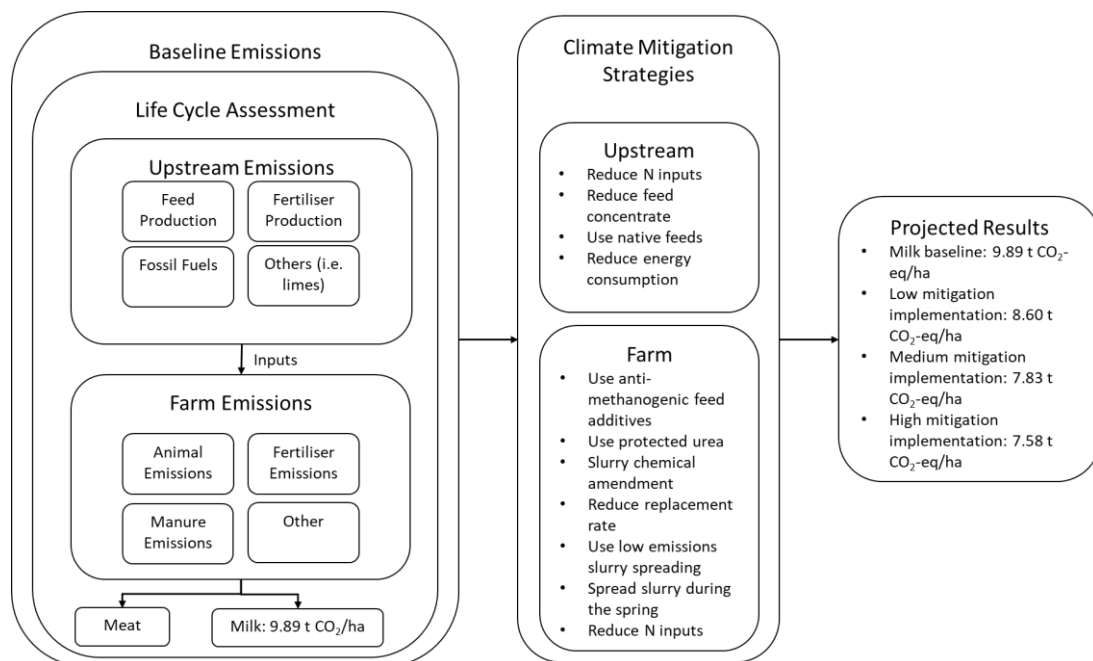
Project Title: The environmental impact of climate mitigation strategies over a commercial Irish dairy farm

Project Leader: Dr. Fionnuala Murphy

Abstract

Agriculture, while being one of the most important industries in Ireland due to job creation, rural development, and economic value, is also the sector with the highest contribution to the country's greenhouse gas emissions. The agricultural system that contributes the most to the sector's emissions is dairy with the impacts increasing over time due to an increase in production driven by the growth of Irish exports. With the country setting a 2030 goal of reducing the emissions from agriculture by 25% when compared to a 2018 baseline, the mitigation of the dairy sector becomes critical in achieving the country climate goals. The development of policy and public incentives to reduce emissions in addition to the implementation of new climate mitigation strategies are required to achieve the required emissions reductions. Farm Zero C is a project that aims to prove that the mitigation of the sector is achievable by implementing climate mitigation strategies at a commercial dairy farm in southwest Ireland. Primary data was collected from the farm in 2018 to perform a life cycle assessment that determined the environmental impacts associated with its milk production. The results showed that the global warming potential of the milk produced in this farm was 9.89 t CO₂-eq/ha. The impacts of implementing a range of climate mitigation strategies were modelled by gathering data from literature. Results show that a low, medium, and high implementation of the strategies would decarbonize the farm to 8.60, 7.83, and 7.58 t CO₂-eq/ha, respectively.

Graphical Abstract



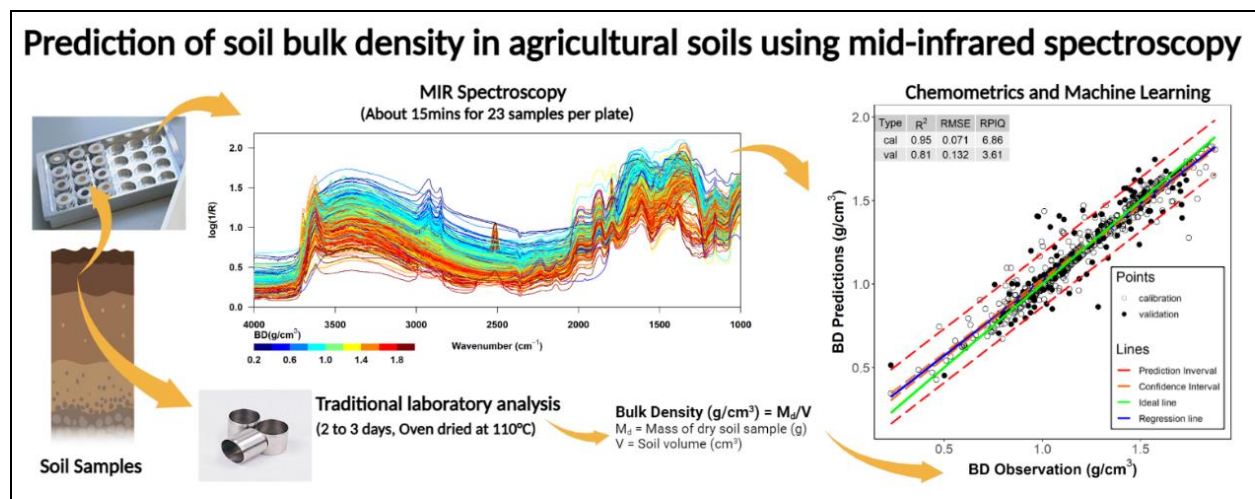
Longnan Shi, PhD student, B.Sc., M.Sc.

Project Title: Predicting soil carbon sequestration potential on Irish soils from spectral data

Project Leader: Assoc. Prof. Sharon O'Rourke

Abstract

This project aims to predict and map soil organic carbon and its sequestration potential on Irish soils, based on national-scale soil spectral libraries with soil chemical and geochemical reference information. With the project going on, three knowledge gaps were identified and fulfilled step by step, which are 1) the missing value of soil bulk density (BD) for carbon stock or sequestration potential calculation; 2) the methodology of estimating soil carbon saturation on Irish soils; 3) how to lead this project to policy-oriented research. Currently, the first knowledge gap was able to be resolved. Soil BD is commonly determined via the core or clod method with laboratory analysis, which is time-consuming, labour intensive and expensive, especially for a national-scale soil assessment. Hence, the omissions of BD values for all or some records are widely found in legacy soil databases. By employing different chemometric and machine learning algorithms, soil BD of Irish soil was able to be estimated from mid-infrared (MIR) spectral libraries by partial least square regression (PLSR), random forest, Cubist and support vector machine (SVM). The best performance was observed for the SVM model with a higher ratio of performance to interquartile distance (RPIQ = 3.61) and R^2 (0.81) values and lower root mean square error of prediction (RMSEP = 0.132). Moreover, compared to the published traditional BD pedo-transfer functions (PTFs), either on overall or specific depth categories, the spectral soil BD model is significantly better rather than traditional PTFs. Hence, high accuracy and the homogeneity of performance of the spectral soil BD model on different depth layers could be noteworthy strengths of spectral modelling techniques when carrying out national soil surveys and large-scale carbon stock assessments.



Selected Recent Publications

Shi, L., O'Rourke, S., de Santana, F.B., Daly, K. (2023) 'Prediction of soil bulk density in agricultural soils using mid-infrared spectroscopy', *Geoderma* 434, 116487.

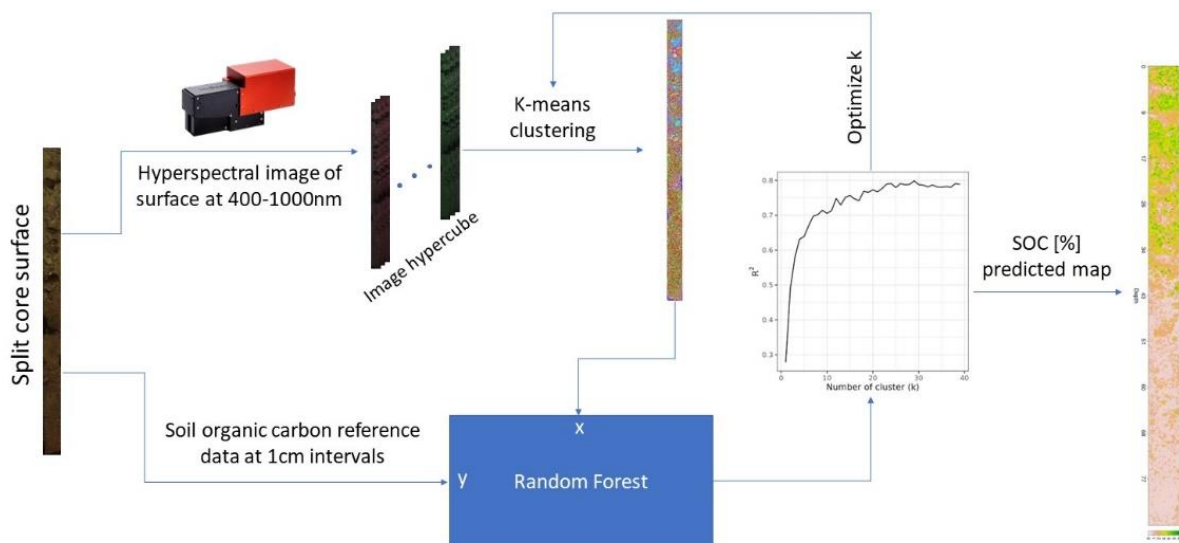
Shayan Kabiri, PhD2

Project Title: Soil organic carbon mapping with semi-supervised learning on Vis-NIR hyperspectral images

Project Leader: Assist. Prof. Sharon O'Rourke

Abstract

Hyperspectral imaging has been an effective tool for modelling and mapping soil properties such as soil organic carbon (SOC), pH, cation exchange capacity, geochemical composition, texture and bulk density. Differences in resolution and dimension between soil images and reference data have been a challenge due to the heterogenous nature of soils. The aim of this study is to develop an SOC model using hyperspectral imaging that addresses the problem with reference samples taken in only one dimension, e.g., in direction of depth. Hyperspectral images taken from the surface of split cores were truncated to a singular unsupervised classified map, and percentages of each class in a 1 cm interval were used as predictors for SOC in that depth interval. The developed model had cross-validated $R^2 = 0.80$ and root-mean-square error (RMSE) = 0.62. This shows combining unsupervised and supervised modelling methods is effective for estimating the distribution of SOC in higher dimension images. Further 2D sampling is needed to assess the exact effectiveness of the method.



EXPLORATION OF GEOPOLRISK TO UNDERSTAND ANTIMONY RESOURCE DEPLETION FOR THE EU THERMOELECTRICS MARKET

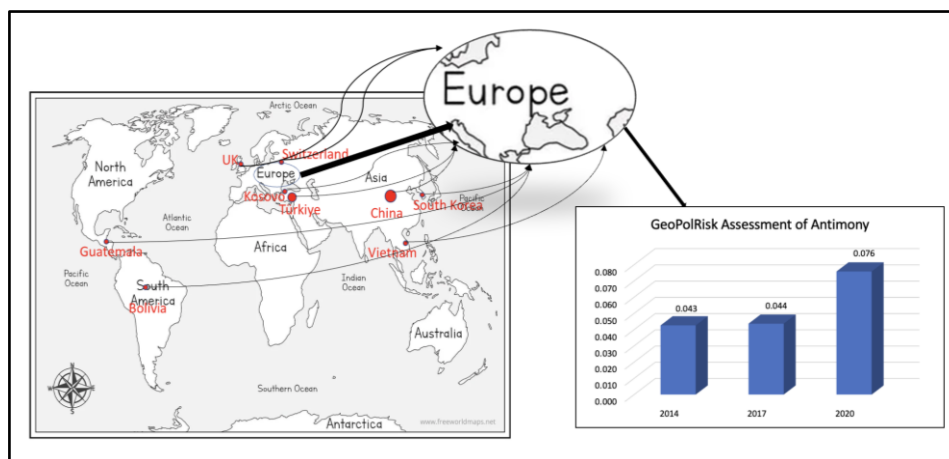
Unza Jamil, Nicholas M. Holden

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Abstract

Antimony is a critical raw material (CRM) used in various applications such as lead-acid batteries, lead alloys, flame retardants, and emerging technologies, such as thermoelectrics. However, antimony deposits are limited, and the current extraction rates could result in depletion of global antimony resources by 2050. Competition for limited resources could have serious implications for the expanding European Union (EU) market for thermoelectric technologies. This paper evaluated the supply risk of antimony in the EU between 2014 and 2020 using the GeoPolRisk method. The GeoPolRisk method quantifies supply risk by considering the global production concentration of the raw material and import shares of trade partners, weighted by their political instability. The data on major importers, total imports, and domestic production were obtained from the EU critical raw materials study for respective years. The GeoPolRisk of antimony imports to the EU increased by about 50% from 2014 to 2020. EU Policies to diversifying supply, increase circularity and research and development into alternative materials and technologies that could replace antimony will be required to ensure a viable market for thermoelectric materials by the middle of the 21st Century.

Graphical Abstract



Introduction

To ensure quality of life, raw resources are crucial. However, competition for limited materials arises with the escalating patterns of production and consumption. The consumption of raw materials is forecast to increase from 79 billion tons in 2011 to 167 billion tons in 2060 (Domaracka *et al* 2022), which brings up significant challenges including resource depletion, price and market instability, the possibility of supply disruption, and other environmental, economic, and social effects (Martins and Castro 2019). The Earth's ability for regeneration is incompatible with forecast rates of raw material exploitation and associated pollution. Future generations are in danger of conflict and lower living standards because of unequal access to resources. At EU level, the Raw Materials Initiative, and the resource efficiency policy both list resource security as a policy goal. Several studies and approaches for evaluating critical raw materials (CRMs) have been created focusing on supply risk and system susceptibility to a

possible disruption of supply. From an economic perspective, supply security is one of the requirements for guaranteeing a steady flow of raw materials (Mancini *et al* 2018).

Antimony is a mineral that is employed in a variety of applications, such as the production of polyester, lead-acid batteries, lead alloys, flame retardants and is an important material for emerging technologies such as thermoelectrics. However, antimony deposits are limited, and it is one of the rarest mineral resources when compared with extractable reserves ($> 2 \times 10^6$ tonnes) and the rate of extraction currently in place (110×10^3 tonnes) (USGS 2023). This rate could result in global antimony resources being exhausted by 2050. Although antimony may not completely vanish, comparatively simple ores may become inaccessible. Due to factors including low ore grades, deep digging, inaccessible areas, and expensive electricity, additional antimony extraction will then become significantly more expensive (Henckens *et al* 2016).

The assessment of the criticality of raw materials aims to present a consolidated view of various risks related to their utilization, whether at a product, company, national, or global level. This process considers several supply risks, which include potential economic, environmental, or social hazards, as well as their possible consequences, often referred to as vulnerability to supply restrictions. Understanding the geopolitical factors is crucial for companies, regions, or countries as it enables them to comprehend the limitations that may affect their supply chain in the short and mid-term, as well as assist them in making informed decisions for their long-term strategy (Gemechu *et al* 2016). It is also important for researchers, as the potential value of new innovations is greatly reduced if they rely on CRMs.

The "Task Force on Mineral Resources" established by the United Nations Environmental Program's Life Cycle Initiative recommends the ESSENZ and GeoPolRisk methods for assessing resource. The ESSENZ method measures accessibility by utilizing a set of indicators that consider socio-economic constraints at a global level, whereas the GeoPolRisk method calculates supply risk at the product level by weighting resource imports based on the political stability of the exporting country. The GeoPolRisk method was initially designed as a midpoint characterization factor for Life Cycle Sustainability Assessment (LCSA), but for this work it was applied independently as a comparative supply risk assessment tool for antimony in reference to EU for the period 2014 -2022 (Koyamparambath *et al* 2022).

The objective of this study was to assess the supply risk of antimony in the context of a growing EU market for thermoelectrics during the period of 2014- 2020 using GeoPolRisk method.

Materials and Methods

GeoPolRisk Method

The GeoPolRisk approach was developed to measure the supply risk of raw materials in a product for a country, region, or group of countries. The method quantifies supply risk by considering the global production concentration of the raw material and the import shares of trade partners, weighted by their political instability. The production concentration is evaluated using the normalized Herfindahl-Hirschman Index (HHI) on a scale of 0 to 1 for raw material extraction or processing, while political instability is estimated using the Political Stability and Absence of Violence dimension of the Worldwide Governance Indicators (WGI-PV). Later, domestic production was incorporated into the GeoPolRisk formula to account for local production of supply requirements. The formula (Equation 1) is:

$$GeoPolRisk = HHI_A \times \sum_i \frac{g_i \times f_{AIC}}{p_{AC} + F_{AC}} \quad (1)$$

where, HHI_A is Herfindahl-Hirschman Index for commodity A, g_i is Geopolitical (in)stability of country i, f_{AIC} is Imports of commodity A from country i to country c, F_{AC} is Total imports of commodity A to country c, p_{AC} is Domestic production of commodity A in country c.

Data Sources

The dimension of Political Stability and Absence of Violence from the Worldwide Governance Indicators (WGI-PV) (Kaufmann *et al.* 2011) is utilized to estimate political instability (g_i) which varies for each country every year. The data on major importers and suppliers (used to calculate F_{AIC} and F_{AC}) for the years 2014 (EC 2014), 2017 (EC 2017) and 2020 (EC 2020) were obtained from EU critical raw materials study for respective years alongside total imports and domestic production (European 2020). The major suppliers to EU kept change over time but China and Vietnam had a share over all the years. For the year 2014 China, Vietnam, Kyrgyzstan, and Russia had 99% share for the total imports (EC 2014). In 2017, 94 % of the total imports were from China and Vietnam (EC 2017). In 2020, the list of importers expanded to China, Korean Rep, Bolivia, UK, Vietnam, Turkey, Guatemala, Switzerland and Kosovo (EC 2020). There was no domestic production of Antimony in the EU (i.e., $p_{AC} = 0$). The data were used to calculate HHI_A for refined antimony import to the EU for 2014, 2017 and 2020.

Results and Discussion

Figure 1 presents the evolution of the geopolitical risks associated with the supply of antimony demand in Europe from 2014 to 2020. It is apparent that antimony supply risk is increasing. GeoPolRisk can be categorized into two primary components, namely production concentration and accessibility. The HHI serves as a measure of production concentration, with a range of values between 0 and 1. A value closer to 0 signifies that production is spread out across multiple countries, while a value of 1 implies that production is centralized in a single country. The latter is linked to the availability of resources to a specific economy. When importing resources from countries with low scores on the WGI-PV indicator of political stability, the risk level may be considered higher. The results depict that across the years the importers have increased with varying production concentration and WGI-PV which takes part in doubling up the GeoPolRisk to 0.076 in 2020.

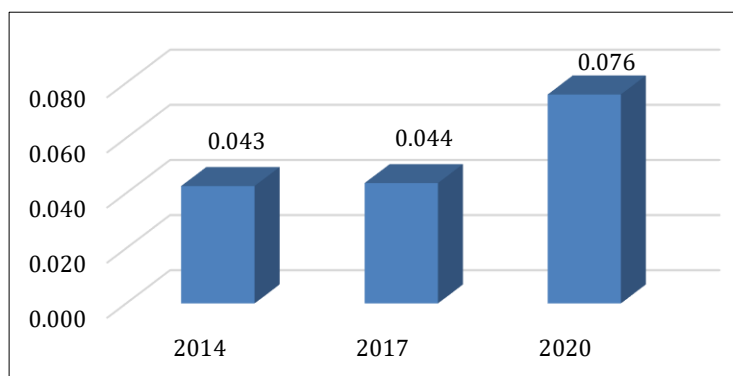


Figure 1. GeoPolRisk Assessment of Antimony from 2014-2020.

In the period studied, Antimony was important for industrial and energy applications such as flame retardants and lead acid batteries, which is likely to continue. However, the emerging technology associated with thermoelectrics is anticipated to drive a \$1.46 billion market by 2033, with a CAGR (capital annual growth rate) of 11.8% between 2023 to 2033 (Pratik M 2021; Newsmantraa 2023). A growth in the thermoelectric market will drive greater demand, which is likely to increase supply risk over the coming 10 to 20 years. If we assume that the HHI, domestic production, and total imports remain the same as in 2020, then any increase in the GeoPolRisk score would be solely due to changes in the geopolitical instability of the supplier countries. Although if the domestic production of antimony doubles in the next 10 years (which would correspond to a CAGR of approximately 7.18%), the new GeoPolRisk score in 2033 would be 0.152. Moreover, the geopolitical instability of the supplier countries increases by 10%, the new GeoPolRisk score in 2033 would be around 0.397.

A significant component of the GeoPolRisk of antimony was because China is the dominant supplier. Dependence on a single supplier makes the EU vulnerable to supply chain disruptions and geopolitical risks associated with antimony because EU itself has no reserves and primary

production (EC 2020). The diversification of suppliers in 2020 is perhaps a response to this risk. GeoPolRisk is estimated based on the imports only however there are several other factors which encompasses criticality of any material such as economic importance, total reserves, increasing prices, trade restrictions, environmental concerns, and low substitutability. Circularity is an option that could increase the value of p_{AC} , which could reduce risk for the EU. It should also be noted antimony production outside the EU effectively ‘exports’ impact on the environment, due to the release of pollutants during mining, processing, and disposal of antimony-containing products. The lack of readily available alternatives makes it challenging to substitute or reduce the use of antimony in many applications, but research and development should focus on this question now, to be ready to mitigate the likely increasing risk by the middle of the 21st century.

Conclusions

Policies aimed at diversifying supply, promoting mineral exploration, and domestic production have become crucial in mitigating the supply risk of raw materials in EU. These measures are observed to effectively reduce the supply risk. The EU has also invested in research and development of alternative materials and technologies that could replace antimony in certain applications. These efforts aim to reduce the supply risk, environmental impact, and economic vulnerability associated with antimony.

Acknowledgements

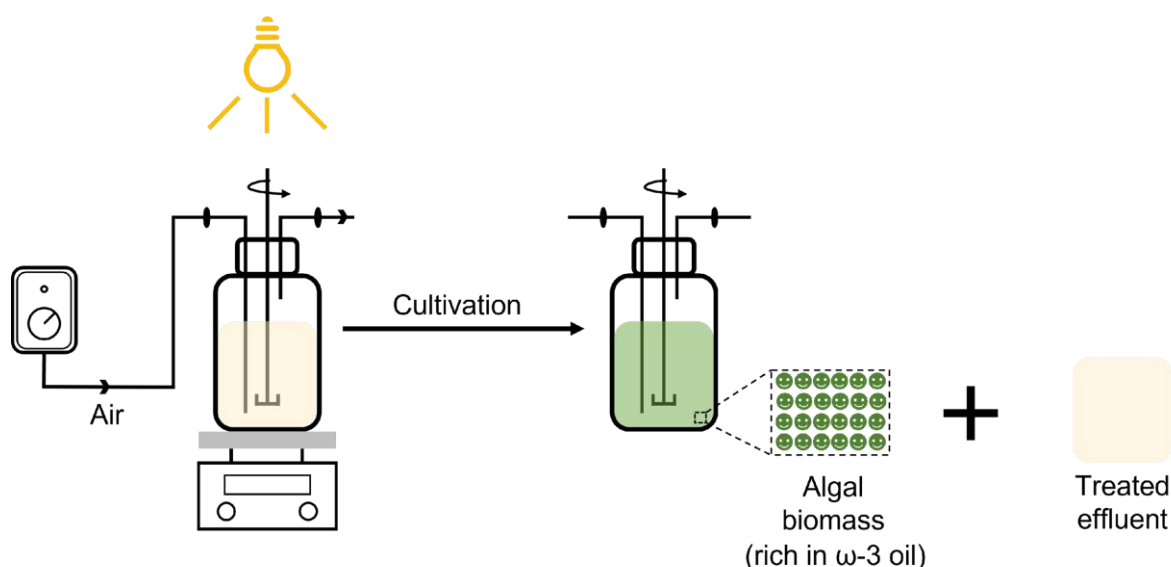
This work was funded by (1) Science Foundation Ireland (SFI) under Grant Number 16/RC/3889 for BiOrbic Bioeconomy, SFI Research Centre, which is co-funded under the European Regional Development Fund and by BiOrbic industry partners; and (2) Atoms-2-Products, Centres for Doctoral Training (CDT) Programme which is a partnership between BiOrbic (Ireland) and University of Nottingham (UK).

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Abstract

This study investigated the application of microalgae cultivation by *Nannochloropsis* species for dairy-wastewater bioremediation and co-production of valuable feed/food ingredients in a circular dairy system (β -galactosidase and omega-3 polyunsaturated fatty acids (PUFAs)). Microalgae can serve as an attractive valorisation strategy for dairy by-products. The results indicated that microalgae can grow on dairy medium by assimilating lactose and soluble proteins and that the system can be effectively applied for onsite dairy wastewater treatment under mixotrophic conditions.



Selected Recent Publications

Kiani, H., Azimi, Y., Li, Y., Mousavi, M., Cara, F., Mulcahy, S., McDonnell, H., Blanco, A., Halim, R., (2023) 'Nitrogen and phosphate removal from dairy processing side-streams by monocultures or consortium of microalgae', *J Biotechnol*, 361, 1-11.

Li, Y., Kiani, H., Eckhardt, H., Blanco, A., Mulcahy, S., McDonnell, H., Tiwari, B.K., Halim, R., (2023) 'Mechanism of lactose assimilation in microalgae for the bioremediation of dairy processing side-streams and co-production of valuable food products' (Manuscript submitted)

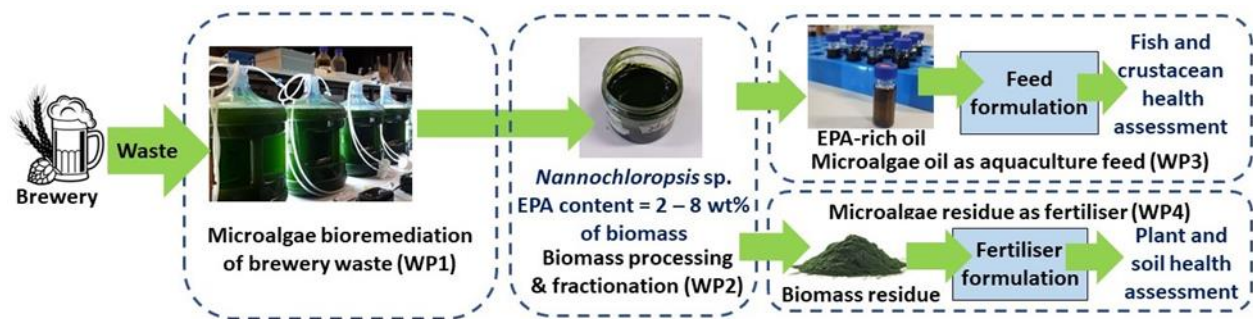
Felix J. Brooke, BSc.

Project Title: Microalgae application of brewery solid waste

Project Leader: Dr. Ronald Halim

Abstract

The brewing industry produces a significant amount of wastewater and solid by-products such as Brewer's spent grain (BSG), which is primarily used as animal feed or sent to landfill. However, researchers are exploring alternative applications for BSG across a broad spectrum of industries to reduce environmental impact and promote sustainable waste management. Microorganisms, including microalgae, can transform BSG into value-added products through cultivation in solid-state and submerged fermentation systems, producing enzymes, lipids for biofuel production, pigments, and other compounds



ECONOMIC SOLUTIONS FOR LIVING OFF THE GRID THROUGH THE INTEGRATION OF VARIOUS RENEWABLE ENERGY SOURCES

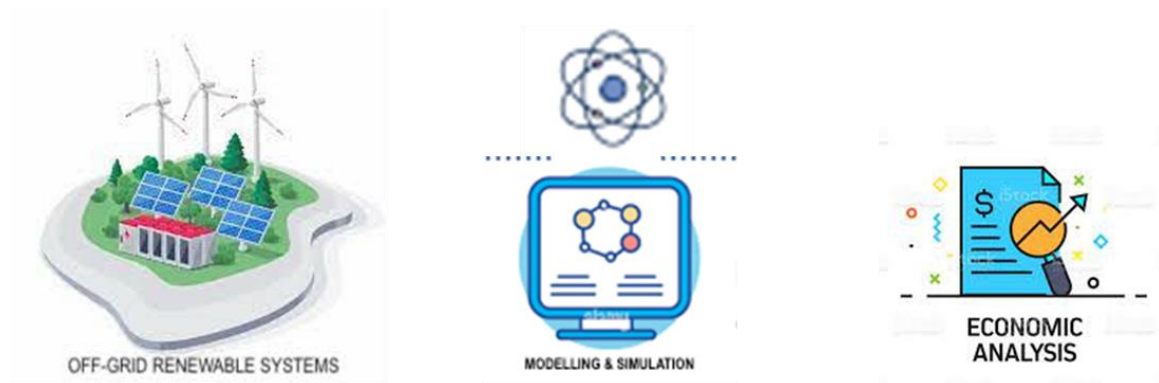
Ofure Iyohaiyoke, Patrick Grace

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

In the journey to a world driven by clean energy, many potential sustainable energy solutions are being explored simultaneously. However, there are still highly vulnerable regions with other energy issues such as, deficit, unreliable energy systems, insufficient capacities, etc. In this research thesis, off-grid renewable solutions are identified as the optimum solution for some developing regions to meet their energy needs, sustainably. This study aims to develop models of technically feasible and economically viable off-grid renewable energy systems that can be adopted to multiple locations to meet their (clean) energy needs. Successful case studies are evaluated to check the feasibility of reproducing the same energy systems in similar locations as the case studies. Software tools such as ArcGIS and HOMER Pro would be used to build the required models and evaluate their technical feasibility and identify the associated financial requirements. Several options for economic solutions would then be explored and recommendations would be given. The expected outcomes of this project are to obtain quantitative estimations of the potential coverage of the off-grid renewable energy systems analyzed, and to suggest feasible economic solutions that would aid the implementation of such projects.

Graphical Abstract



Introduction

Despite various efforts and the promising trajectory of the energy sector, there is still a huge gap to be filled to reduce energy poverty and produce cleaner forms of energy. The current energy deficit is about 770 million of the population, predominantly in Sub-Saharan Africa and South Asia. This number is expected to reach 1 billion by 2030, owing to several factors including population growth (IEA 2022). Apart from lack of access to energy, the areas primarily affected also face other energy issues energy such as: inconsistent availability, lack of clean energy alternatives, and grid failures. In 2022, Nigeria, with over 40% of its population without energy access, experienced nationwide blackouts from grid failure at least six times (Udegbumam 2022).

With growing technology and investments in renewables, more options are available for solving the current energy needs. The World Economic Forum posits that renewable technologies can potentially meet the energy needs of 70% of the population without energy access, by 2030. It was further stated that off-grid renewables can cover 50% of the renewable solutions. This means off-grid renewable energy solutions can fill the gap of energy access for 35% of the population by 2030 (Daly 2018). This is a significant number worth exploring to discover available options.

Beyond the technical feasibility of the promising solutions which can potentially make a significant difference, there are still other hurdles to be overcome by the vulnerable regions who need these solutions the most. The developing countries that could benefit from these solutions lack the financial capacities and supporting policies to drive projects in this regard (Ngowi *et al* 2019).

Based on the background highlighted, and considering the numerous options being explored to cater to the various energy issues, this thesis project is focused on the ‘Economic solutions for living off the grid through the integration of various renewable energy sources’. This would be carried out based on two major goals.

- To identify and develop profiles for implementing similar renewable energy systems. This means that successfully implemented renewable energy projects in developing countries could be examined. Then, profiles of similar locations would be developed, to evaluate the possibility of obtaining similar results if the successfully implemented projects are replicated.
- To propose feasible economic solutions for off-grid renewable technologies. This would be achieved by first identifying the major financial limitations that the developing countries experience regarding implementing renewable energy projects. Then, cues would be gotten from global references, as well as from countries that were able to find sustainable financial solutions. From these, feasible economic solutions would be recommended for implementation.

The objective of this project is to develop replicable ‘models’ of off-grid renewable energy systems for locations with high energy deficit, with feasible economic solutions.

Material and Methods

This project would involve a quantitative assessment and would include several case studies. The materials needed are briefly discussed in relation to the step-by-step methodology, depicted in Figure 1.



Figure 1. Methodology approach

- *Literature.* The material that would be used here are the case studies that have been assessed or implemented, with positive conclusions. This would involve assessing previous research works that have studied successfully implemented renewable energy system projects. (Vendoti *et al* 2021) explored different Hybrid Renewable Energy Systems (HRES) for off-grid rural electrification in villages in India, with positive conclusions. (Shahzad *et al* 2017) analyzed a solar-biomass off-grid system in rural remote areas in Pakistan and concluded that government-supported financial incentives would ensure easier implementation of these systems. These case studies are referred to as ‘profiles’, which serve as the starting point for the research project. Data on population dynamics, demography, energy usage, available renewables, and economic activities would be obtained from previous literature and research works, specific government agencies, energy organizations, and statistical bodies.
- *Geographical mapping.* Here, the material to be used is the Geographical Information System software, ArcGIS. This would be used to identify areas that would fit into profiles that have been described from literature. From the case studies highlighted earlier, the ArcGIS software would be used to map out other locations like the rural villages in India, and the remote areas in Pakistan. This geographical mapping would be based on similarities like topography, solar irradiation, wind frequencies, presence of hydro, etc.
- *Modelling & Simulation.* The material used in this step of the methodology is the HOMER Pro software. HOMER stands for Hybrid Optimization of Multiple Electric Renewables and is applicable in this thesis project for designing, optimizing, and carrying out sensitivity analysis on proposed energy systems. HOMER Pro would be used to evaluate energy systems of profiles developed, based on similarities with case studies already conducted in literature. Reiterating the first two steps, HOMER Pro would be used to model the same energy systems from the case studies, but in the locations obtained from geographical mapping. The aim is to compare results to identify similarities that would confirm the practicality of replicating these energy systems.
- *Analysis.* This begins the discussion of results and involves examination of the data obtained from HOMER Pro, as well as comparison with data obtainable from previous case studies. The results of the energy systems modelled in HOMER Pro for the locations obtained using ArcGIS would be compared with those in the case studies referenced in literature. The degree of similarity in the outputs in terms of finances and electricity generation would be an indicator to show the feasibility of replicating these energy systems in other similar locations. This step of the methodology would cover the sensitivity analysis from HOMER Pro, and economic analysis to address funding options, loans, pay-back period, to identify feasible replicable financial solutions for funding renewable energy off-grid projects.
- *Recommendations.* From the analysis carried out, recommendations would be made based on the options evaluated. These recommendations would address issues relating to funding, supporting policies and investment alternatives that would be best suited for the regions that are mostly in need of off-grid renewable systems.

A simplified summary of the methodology is given below.

- A case study is identified from previous research work. The case study was on a hybrid renewable energy system (solar-biomass) in a rural community in South Asia.
- A similar location is identified in Sub-Saharan Africa using ArcGIS.
- HOMER Pro is used to model a hybrid renewable energy system (solar-biomass) for the location in Sub-Saharan Africa.
- Analysis is carried out based on the results obtained, with reference to those of the case study.
- Recommendations are made and conclusions drawn.

Results and Discussion

This project is at the very early stages of development. Currently, there are no research works which can be referenced with the same aim and objectives as this thesis. Therefore, the quantitative results cannot be projected at this stage. However, based on the case studies evaluated and the proposed methodology, the following are expected to be achieved at the end of the project thesis:

- Successful replication of modelled scenarios in identified profiles. It is expected that the systems that would be modelled in HOMER would give similar results to those identified in case studies.
- Feasible economic recommendations. At least 3 replicable financial solutions to be given based on the analysis carried out.
- It is expected that the thesis would propose an estimate of the potential population that can be covered by the implementation of the replicable models. Although not all the locations identified using ArcGIS would be modelled in HOMER Pro, the data obtained from this location would be used to give an estimate of the potential reduction in energy deficit and access to clean energy that is obtainable with the implementation of these off-grid renewable energy systems and adoption of the recommended economic solutions in multiple locations.

The limitations encountered during this research project would be fully highlighted and discussed to serve as a basis for further research to be carried out on this topic.

Conclusion

The potential impact of this research topic would make a significant contribution toward accomplishing the seventh of the SDGs (affordable and clean energy), by 2030. By identifying economic solutions for implementing renewable energy systems in highly vulnerable areas, we would get closer to achieving global access to energy, and an increase in renewable energy coverage.

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LITERATURE AND MARKET REVIEW OF MYCELIUM-BASED BIOCOMPOSITE MATERIAL

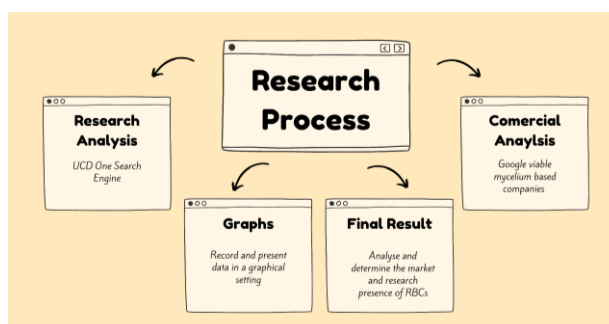
Michelle A. Finneran, Dimitrios Argyropoulos.

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Abstract

Mycelium-based biocomposites have garnered more attention in recent years for its environmentally friendly applications in the packaging, design, and construction industries. The potential for this material to supplement polystyrene and other non-renewable materials with its biodegradable and lightweight characteristics shows promise and drives modern research. As more focus falls onto mycelium, more innovation and technological improvements will follow. This paper presents a literature review of the current commercial and scientific fundamentals to detail how mycelium is used in the current market and outline barriers to be addressed for further implementation. The results of the review showed that mycelium is mostly introduced through the food and packaging sectors and problems with the material's density and water absorbance need to be resolved to further penetrate the market. Although mycelium research is still in its infancy there is shown to be an increase in research interest for this material.

Graphical Abstract



Introduction

With fossil fuels being an unsustainable resource there arises a strong desire to shift towards more sustainable methods for material manufacturing. Recently mycelium has risen as a dynamic material that shows great potential as a sustainable alternative to many products. Mycelium is considered to be the vegetative rootlike structure of the fungi, the individual tubular filaments of the mycelium are called hyphae and its elongated cell wall is what provides structure and its mechanical properties to the material (Haneef *et al* 2017). When grown off of varying substrate it can create a composite structure that is lightweight and presents varying densities that have a variety of potential uses such as packaging foam, vegan leather, and construction panels (Girometta *et al* 2019). Mycelium based bio-composites (MBC) come from the spawned fungal spores that grow and fully bind to some organic matter substrate (Appels *et al* 2019). Much of the sustainability surrounding mycelium has been due to its ability to valorise bio waste streams, producing a material that is lightweight, flame resistant, biodegradable, and presents good insulating properties (Jones *et al* 2018). Due to its flexible nature *Pleurotus ostreatus* is often used in MBC research as it grows off a wide range of organic waste such as fibers, husks, and other lignocellulosic matter (Girometta *et al* 2019). Once fully grown, a novel bio-based product is made that has been used within the fashion, design, architecture, construction, and packaging industry. More developed research examines how mycelium is potentially a sustainable alternative for the packaging industry; even a direct replacement for

the environmentally hazardous polystyrene (Girometta *et al* 2019). However, improvements such as reducing the material's water absorptivity trait and increasing the mycelium biocomposite's tensile strength are both needed to be addressed in order for a one to one replacement can happen within the industry (Irbe *et al* 2022).

The objective of this study was to develop an understanding of the development of mycelium-based biocomposites in order to identify current research gaps.

Materials and Methods

The main methods used for the literature review in this report was utilizing Scopus that was accessed via the University College Dublin's one search engine. Keywords such as *mycelium* and *mycelium biocomposite* were used when conducting the literature review along with using the UCD search engine. The main method used for the market review was web searches. Keywords such as *mycelium-based companies*, *mycelium products*, and *mycelium biocomposites* were used when doing the commercial analysis.

Results and Discussion

Literature analysis

This paper examined research surrounding mycelium biocomposites, where a total of 54 journal articles were identified. Publication date, research focus, substrates, and fungal type were all recorded and are presented visually below.

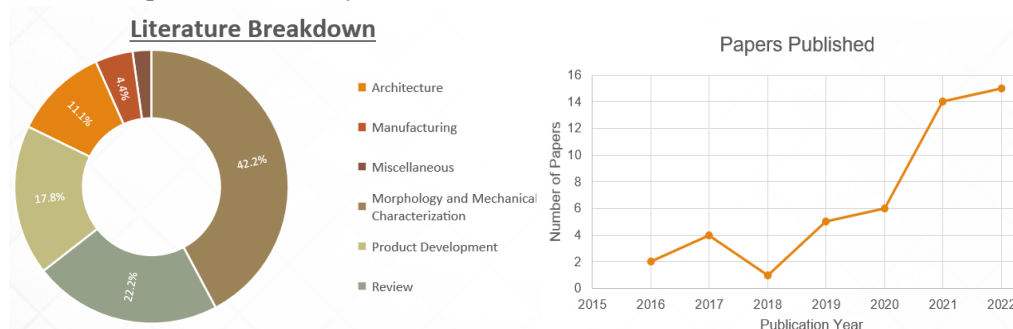


Figure 1. (a) Literature breakdown of the papers digested for this study (b) Timeframe each research journal has been published.

As seen in Figure 1b above there has been a large increase in publications of mycelium research around 2020. The earliest recorded publication was published in 2016. Overall Figure 1b shows the novelty of the material and how the subject matter is starting to gain momentum and support from within the scientific community. Of the research being done Figure 1a shows that over 42% of papers were focused on testing and identifying the morphology and mechanical properties of mycelium. Within this field it was found that the general focus of these papers sought to vary the substrate composition and/or the fungal type and analyze the MBC using scanning electron microscope, mechanical strain tests, moisture content/density analysis. The next largest group of research papers, at 22.2%, were focused on conducting a general review on the potential for mycelium biocomposites and their role to supplement more environmentally damaging materials. These reviews often relied on the results from the characterization studies and would then surmise potential uses for the material within commercial and public sectors. The third largest focus of mycelium research, around 17.8% of journal articles, were based in product development. These papers focused mostly on developing the biobased material as an alternative to polystyrene in the packaging industry, noting its uniquely similar characteristics to the material with the added benefit of its environmental sustainability. Within this grouping of papers there was heightened interest in developing a mycelium based sandwich structure, a design that improved on the mechanical strength and support of the biomaterial as opposed to organically letting the biomaterial form (Jiang *et al* 2017). This design was often examined as a means to test out mycelium in the textile and construction industry as a potential bio leather or

paneling for construction material. Further breaking down the literature, the papers analyzed the most common substrate and fungal strain were recorded and can be seen in Figure 2 below.

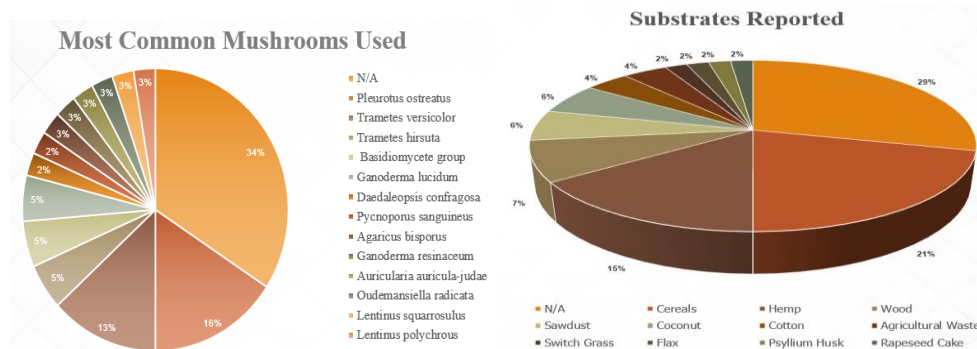


Figure 2. (a) Breakdown of the different types of mushrooms used in the literature (b) Breakdown of the substrate types used in the literature.

As seen above, the most common mushroom used was not specified; however the next most common mushroom used in these experiments was the *Pleurotus ostreatus* followed by the *Trametes versicolor*, and *Trametes hirsuta*. The heavy use of *P. ostreatus* is due to its flexibility in being able to grow off of a wide range of substrates (Hoa *et al* 2016). When looking at the different substrates used often the substrate makeup wasn't recorded in the paper, however when it was it was most often a cereal at 21%, then hemp at 15%, then the third most was a wood substrate at 7% of the publications. Generally it was found that much of the research was set within a laboratory environment and the main focus of the research was to determine how changing the fungal type and substrates affected the properties of the biocomposite material.

Commercial analysis

Along with a literature review, research was conducted to determine new companies that utilized the novel mycelium biocomposite material. The below Figure 3 shows the timeline of startup companies that have a central focus on mycelium as their product. Of the 36 companies found over 40% were focused on utilizing mycelium within the food sector (Figure 3), the next most common start up at 26% looked at mycelium as a packaging material, and the third most (reported at 17%) focused on utilizing mycelium within the fashion industry. The majority of these companies were based in the United States at around 36% with the next largest country being the Netherlands, housing around 17% of the companies. The initial start-up dates were tracked and represented below in Figure 3.

These results show an increase in market interest in the practical uses of mycelium as a material. Figure 3 shows a spike in startups around 2019 which corresponds to the reported increase in papers published around 2020 in Figure 1b. Within the packaging industry companies like Ecovative are propelling the material into mainstream markets as they have partnered with brands such as Dell and Calvin Klein for their packaging and mycelium leather services (Ecovative, 2022).

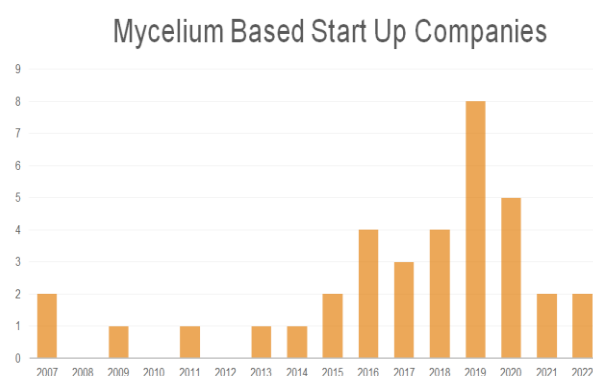


Figure 3. Timeframe of mycelium based companies formed.

Conclusions

Much of the research done with mycelium as a bio-composite is still in its early phases. The majority of research is focused on small scale in laboratory experimentation where the main goal focuses on discovering how changing the fungal type and substrate affect the morphology and characteristics of the MBC. The majority of the publications were exploratory and all used small batches in a sterile laboratory environment when examining the material. There is almost no research being done with large batches or in a field type setting. However, there is a strong emphasis on mycelium as a potential to supplement a variety of manufactured fossil-based materials. From this more research is clearly needed to fully extract a greater range of value from this promising material as the research is still in its infancy. Some of the prevailing questions left suggest several hurdles that will need to be overcome such as what substrate and fungal strain make for the best mechanical properties of the biomaterial.

Acknowledgements

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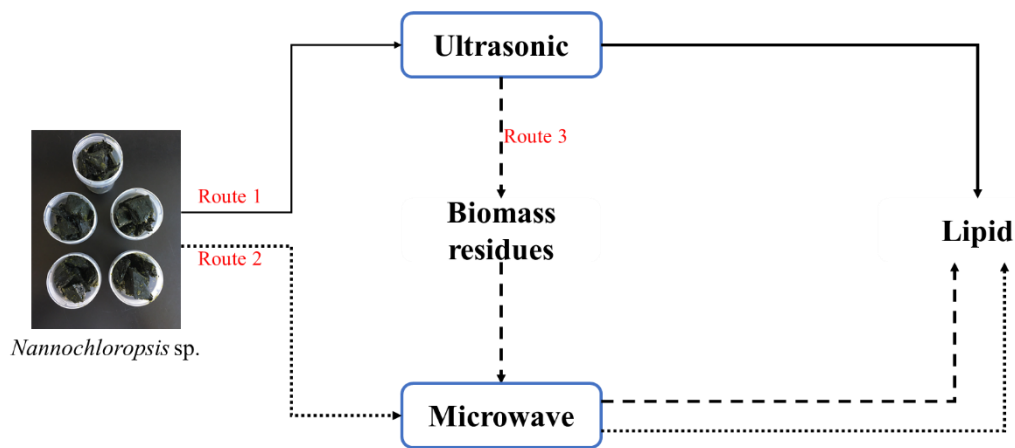
Mohammadhosein Rahimi, BSc, MSc.

Project Title: Lipid extraction from wet microalgae biomass for the production of omega-3 rich animal feed

Project Leader: Dr. Ronald Halim

Abstract

Nannochloropsis sp. are among microalgae species with high concentrations of functional lipids, which has attracted considerable attention worldwide. However, these species' cell wall rigidity is a challenge in extracting the lipids. In this study, the lipid content of wet microalgae *Nannochloropsis* sp. was determined using ultrasonic and microwave-assisted methods. Two different solvent mixtures were used during the extraction process. The highest lipid extraction yield (85.1 mg/g biomass) was obtained by performing a two-stage extraction using a mixture of chloroform and methanol.



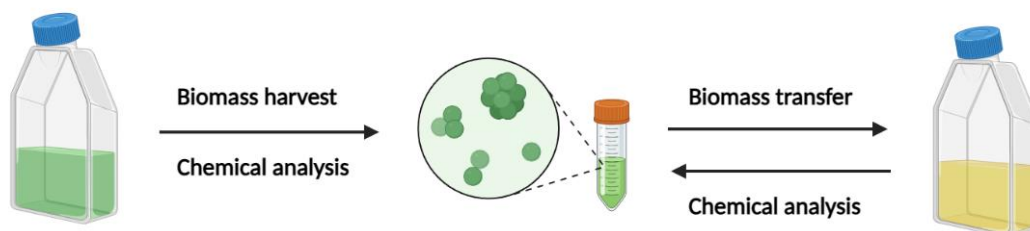
Mengsong Xiao, BSc, MSc.

Project Title: Microalgae bioremediation of dairy wastewater

Project Leader: Dr. Ronald Halim

Abstract

In this study, the effect of microalgae bioremediation of dairy wastewater was investigated through the cultivation of *Tetradesmus obliquus*. The microalgae was grown in nano-filtered whey permeate and standard media for seven days, and then the cells were harvested. The optical density of the culture grown on the whey permeate sample was significantly higher than that of the control, and the dry biomass concentration of the culture on whey permeate and control sample were found to be 0.56 and 0.01 g/L, respectively. We hypothesize that multi-stage cultivation of microalgae can produce microalgal biomass and remove contaminants more effectively than a single-stage cultivation.



ENHANCING FOOD SAFETY OF FRESH POULTRY MEAT USING COLD PLASMA TECHNOLOGY AND NATURAL COMPOUNDS

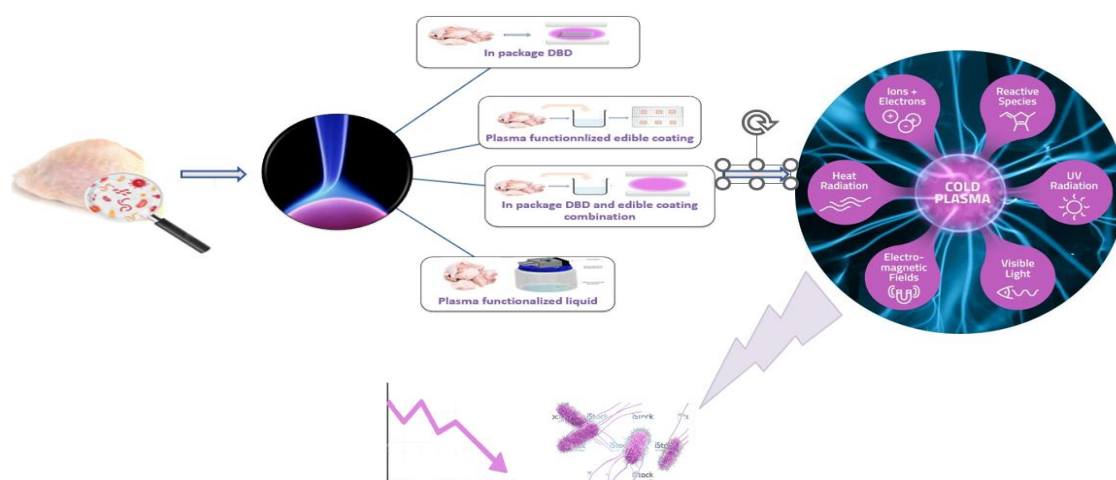
Schehrazad Bey, Paula Bourke

UCD School of Biosystems and Food Engineering, University College Dublin, Belfield, Dublin 4, Ireland.

Abstract

The aim of this study was to evaluate the antimicrobial effect of three approaches of cold plasma application (In package treatment, Plasma functionalized liquid and Plasma functionalized edible coating) against background microflora in fresh poultry meat, separately and in combinations, and then assessing the effect of combined treatments on extending shelf-life in terms of quality and microbial safety. Ten different treatments were conducted and the results showed that the combination of 1-minute treatment with dielectric barrier discharge (DBD) plasma and chitosan with lemongrass essential oil achieved the highest reduction of microbial load. This study suggests that the use of cold plasma technology with chitosan and lemongrass essential oil has the potential to enhance food safety. Further studies are needed to determine the optimal conditions for this treatment and to assess its impact on the shelf life and sensory quality of fresh poultry meat.

Graphical Abstract



Introduction

Poultry meat is an important source of protein and nutrients for human consumption worldwide. According to the Food and Agriculture Organization of the United Nations (FAO), in 2020, global poultry meat production reached a record high of 97.1 million tons, making it the largest meat-producing sector. However, it is also one of the main sources of foodborne diseases, including salmonellosis and campylobacteriosis (Naskar *et al* 2017). To prevent the growth of spoilage and pathogenic bacteria, various methods have been developed, including the use of chemical preservatives. However, these traditional methods have raised concerns about their safety and sustainability due to the potential formation of harmful by-products and environmental impacts (Liu *et al* 2021). Therefore, there is an urgent need for safe and sustainable ways to preserve the quality and safety of chicken meat.

One of the promising approaches that have emerged in recent years, is the use of edible coatings, which can protect the chicken meat from physical, chemical, and microbial damages during

storage (Zheng *et al* 2019). Edible coatings are usually composed of natural polymers such as chitosan, which has antimicrobial properties and can act as a barrier against oxygen and moisture. Moreover, the incorporation of natural antimicrobial agents, such as lemongrass essential oil, into the coating can enhance its effectiveness in controlling microbial growth and extending the shelf life of the coated chicken (Lai *et al* 2018).

Another promising eco-friendly method for improving the safety and quality of poultry meat is cold plasma (Zhang *et al* 2021). Cold plasma generates a variety of reactive species including ozone, hydrogen peroxide, and radicals, that can modify the surface properties of food products and induce oxidative stress in microorganisms, leading to their inactivation (Bourke *et al* 2017). Additionally, it enhances the antimicrobial efficacy of edible coatings including chitosan.

This study aims to investigate the antimicrobial effect of three approaches of cold plasma application (In package treatment, Plasma functionalized liquid and Plasma functionalized edible coating) against background microflora in fresh poultry meat, separately and in combination, and then assess the effect of combined treatments on extending shelf-life in terms of quality and microbial safety.

Materials and Methods

Samples preparation

The coating solution was prepared according to a modified method based on previous studies (Wang *et al* 2021). Chicken thighs were cut into 10g pieces. Chitosan powder was treated with the dielectric barrier discharge (“DBD” system for 1 minute and left in the sealed box for 24 hrs to retain the treatment effect. The powder was then dissolved in acetic acid to prepare the Chitosan solution (2% w/v) and the solution was left to stir and heat at 55°C overnight. Following that, Lemongrass essential oil (0.4% v/v) was added to the Chitosan solution to prepare the Chitosan + lemongrass coating solution. The chicken pieces were then dipped in the obtained coating solution for two minutes to create a uniform layer on the surface and left to dry for 20 min afterwards.

The coated chicken samples were then analyzed for quality and microbial safety characteristics after 1 hrs and 24 hrs storage time.

Treatments

Table 1 provides a summary of the different treatments including in-package DBD (Dielectric Barrier Discharge) plasma treatments applied to chitosan powder and/or coated samples. The treatments are listed in the first column, while the second and third columns indicate whether the treatment involved in-package DBD treatment for chitosan powder or coated samples, respectively.

Sample Analysis

Microbial safety

Microbial analysis was conducted on the samples after 1 hour and 24 hours of storage at refrigeration temperature (4 °C). The chicken samples, weighing approximately 10g, were homogenized with 90mL of Maximum Recovery Diluent (MRD) for 2 min to prepare a 1 in 10 dilution for sample suspensions.

For the determination of total viable counts and psychrotrophs, appropriate dilutions were made, and 100 µL of the sample suspension was spread on the surface of Tryptic Soy Agar (TSA) plates. The TSA plates were then incubated at 37°C for 24 hours for total viable counts and at 4°C for 7- 10 days for the psychrotrophs. For the Enterobacteriaceae, the sandwich method was used, which involves adding 100 µl of the sample suspension to a Petri dish followed by adding a layer of Tryptic Soy Yeast Extract (TSY), and then a second layer of Violet Red Bile Agar (VRBA) on top of the first layer. The VRBA layer provides the selective

medium for Enterobacteriaceae, and the TSY layer provides the nutrients for bacterial growth. The plates were then incubated for 24h at 37°C. After incubation, the bacterial colonies were counted, and the results were expressed as log₁₀ CFU/g. All samples were plated in duplicate. All experiments were performed at four replicates.

Table 1. Summary of Treatments and In-Package DBD Plasma Treatment for Chitosan Powder and Coated Samples.

Treatment	In package DBD for chitosan powder	In package DBD for coated samples
1. Untreated Control	/	/
2. Treated control (chitosan)	/	/
3. 1min DBD Chitosan	1 min	/
4. Chitosan + 1min in package DBD	/	1 min
5. Chitosan + 3min in package DBD	/	3 min
6. Chitosan + Lemongrass EO	/	/
7. Chitosan + Lemongrass EO+ 1 min in package DBD	/	1 min
8. Chitosan + Lemongrass EO+ 3 min in package DBD	/	3 min
9. 1 min DBD chitosan + 1min in package DBD	1 min	1 min
10. 1 min DBD chitosan + 3 min In Package DBD	1 min	3 min
11. 1 min DBD chitosan + Lemongrass EO	1 min	/

Data analysis

In this study, the data analysis methods utilized a combination of graphical and statistical techniques. The mean and standard deviation of microbial counts for each treatment group were plotted using bar graphs, enabling to visualize any differences or trends between groups. Statistical analysis was performed using SPSS software, with ANOVA used to determine any statistically significant differences in the microbial counts between treatment groups. The least significant difference (LSD) test was used for post-hoc comparisons between the treatment groups, enabling identification of significant differences compared to the control group. The combination of graphical and statistical methods allowed for a comprehensive data analysis, yielding meaningful conclusions on the impact of the different treatments on the microbial safety of the poultry.

Results

Table 2 shows an overview of the results of this study, which demonstrate the effectiveness of DBD plasma treatment combined with chitosan and lemongrass essential oil (EO) as an efficient plasma functionalized edible coating for chicken. Among the ten treatments tested, the 1min DBD treated chitosan combined with lemongrass EO treatment showed the highest microbial reduction for total viable count (2.53 log), Enterobacteriaceae (1.38 log), and psychrotrophs (2.1 log) at both day 0 and day 1. These findings indicate LEO may have synergistic antimicrobial effects in combination with the chitosan that have been treated for 1 min with DBD, the Cold plasma treatment introduced reactive species that enhanced its efficacy compared with 0 min chitosan treated control. The results also indicate that treatment time is a critical factor in optimizing the antimicrobial activity of DBD in package treatment and that longer treatment times may not necessarily lead to greater reductions in microbial load. The combination of DBD plasma treatment with chitosan and lemongrass EO could provide a novel approach to reducing microbial growth in food packaging, which could enhance food safety, while reducing reliance on traditional preservatives.

Table 2. The effect of each treatment on the TVC, Enterobacteriaceae and psychrotrophic counts on fresh chicken

Parameters	Storage Time (h)	Untreated control	Treated control (0min chitosan)	0 min DBD Chitosan + 1min DBD	0 min DBD Chitosan + 3 min DBD	0min DBD chitosan + LG	0 min DBD chitosan + LG + 1 min	0 min DBD chitosan + LG + 3 min	1 min DBD Chitosan	1min DBD chitosan + 1min DBD	1min DBD chitosan + 3min DBD	1min DBD chitosan + LG
TVC	1	4.69±0.21	4.15±0.23	3.99±0.51	4.38±0.71	3.97±0.15	3.89±0.27	3.51±0.23	3.48±0.42	3.49±0.29	3.59±0.64	2.34±0.36
	24	5.64±0.17	4.28±0.31	4.34±0.44	4.12±0.69	4.43±0.33	4.25±0.64	4.17±0.25	4.04±0.38	4.17±0.41	4.21±0.62	2.93±0.17
Enterobacteriaceae	1	3.78±0.27	2.96±0.20	3.12±0.44	3.14±0.39	2.41±0.41	2.30±0.34	2.13±0.19	2.68±0.46	2.74±0.23	2.83±0.57	2.36±0.32
	24	3.98±0.11	3.31±0.21	3.51±0.44	3.22±0.61	2.65±0.27	2.91±0.45	3.02±0.18	3.40±0.21	2.90±0.35	3.32±0.50	2.64±0.19
Psychrotrophes	1	4.93±0.17	4.26±0.30	4.27±0.08	5.13±0.09	4.40±0.21	4.27±0.28	4.05±0.41	3.88±0.29	4.68±0.25	4.12±0.65	3.42±0.38
	24	5.84±0.06	4.82±0.22	5.10±0.28	4.14±0.58	4.59±0.29	4.73±0.43	4.59±0.24	4.49±0.33	4.69±0.12	4.66±0.37	3.15±0.21

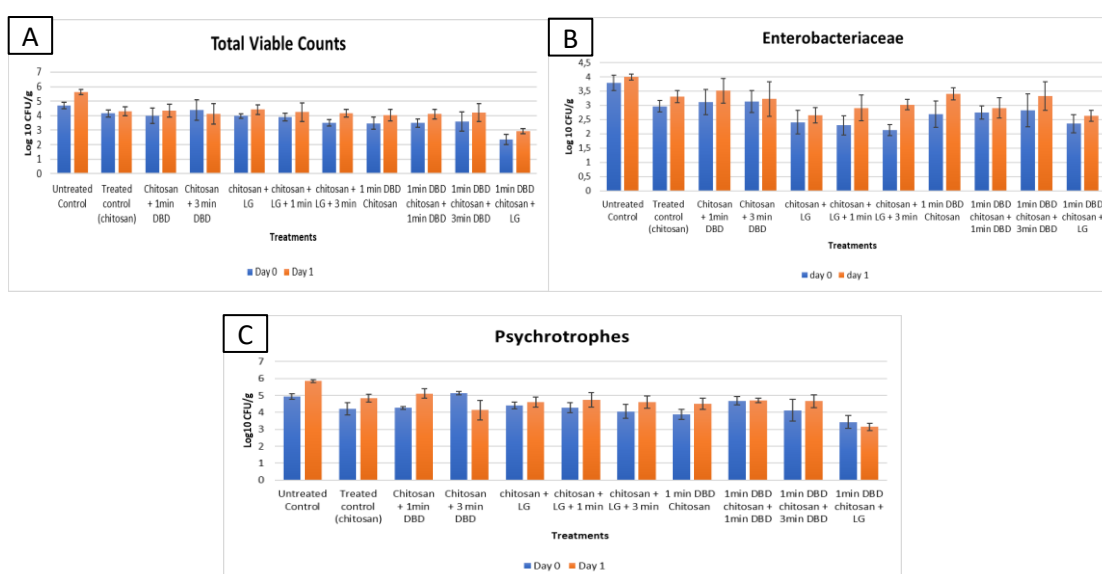


Figure 1. The effect of treatments on the survival of TVC (A), Enterobacteriaceae (B) and psychrotrophs (C) on fresh chicken

Conclusion

In conclusion, the use of cold plasma technology in combination with chitosan and lemongrass essential oil has shown promising results in enhancing food safety of fresh poultry meat. Further experiments are needed to determine the optimal treatment time and concentration of the coating to achieve maximum reduction of microbial load. Additionally, a shelf-life study is necessary to assess the quality of the treated chicken over time. Overall, this study provides a foundation for further investigation into the use of cold plasma technology with chitosan and lemongrass essential oil as a potential method for improving food safety in the fresh poultry industry.

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Appendix A

Award winners for excellence in Research Presentation 2023

F4S 2023 Bai Fangting

Session: Environmental technology/modelling/risk assessment 1

Comparison of portable instrumentation for hyperspectral imaging of bacteria

F4S 2023 Bey Schehrazed

Session: Sustainable Energy/Green Technology

Enhancing food safety of fresh poultry meat using cold plasma technology and natural compounds

TM 2023 Menon Shruti Venugopal

Session: Food Engineering 1

Influence of cold plasma on protein enriched wheat flour

TM 2023 Ravichandran Priyadharshini

Session: Food Engineering 3

Co-cultivation of microalgae and fungi, focusing on microalgae cell walls

Junior PhD 2023 Oliveira Gama Ferreira Raphaela

Session: Imaging/Risk Assessment/Biomedical

Exploring characteristics of microplastics contributing to the cytotoxicity of Caco-2 cells

Junior PhD 2023 Brooke Felix Joel

Session: Sustainable Energy/Green Technology

Microalgae application of brewery solid waste

Senior PhD 2023 Shi Longnan

Session: LCA/Sustainable Agriculture & Soil Resources

Predicting soil carbon sequestration potential on Irish soils from spectral data

Senior PhD 2023 Charisis Christos

Session: Environmental technology/modelling/risk assessment 1

Deep learning techniques for yield prediction in multi-domain mushroom production environments

Senior PhD 2023 Chaple Sonal Ganpat

Session: Food Engineering 1

The effects of cold plasma treatment on physicochemical & rheological modification of hydrocolloids

Senior PhD 2023 Zhu Xianglu

Session: Food Engineering 3

Hydrodynamic cavitation for brown seaweed in a cascading biorefinery model for laminarin, alginate and protein extraction

