



2026

DPTC Publications

DPTC Publications from Core and
Externally-Funded Research

Book of Publications

Acknowledgements

We would like to thank all Principal Investigators, researchers, and industry partners for their collaboration and hard work, which have resulted in these 225 peer-reviewed publications. Your contributions have been essential in advancing the DPTC research agenda.

We appreciate the support from our industry collaborators, which has greatly enhanced our research. To our researchers, your dedication and efforts are the driving force behind our achievements.

This book of abstracts reflects the collective success of our community. Thank you for your ongoing commitment.

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DPTC 2 Publications: RFA 1

Funding Source: DPTC 2

Research Programme: RFA1

Colorants in cheese manufacture: Production, chemistry, interactions, and regulation

Sharma P.; Segat A.; Kelly A.L.; Sheehan J.J.

Comprehensive Reviews in Food Science and Food Safety, 2019

Colored Cheddar cheeses are prepared by adding an aqueous annatto extract (norixin) to cheese milk; however, a considerable proportion (~20%) of such colorant is transferred to whey, which can limit the end use applications of whey products. Different geographical regions have adopted various strategies for handling whey derived from colored cheeses production. For example, in the United States, whey products are treated with oxidizing agents such as hydrogen peroxide and benzoyl peroxide to obtain white and colorless spray-dried products; however, chemical bleaching of whey is prohibited in Europe and China. Fundamental studies have focused on understanding the interactions between colorants molecules and various components of cheese. In addition, the selective delivery of colorants to the cheese curd through approaches such as encapsulated norixin and microcapsules of bixin or use of alternative colorants, including fat-soluble/emulsified versions of annatto or beta-carotene, has been studied. This review provides a critical analysis of pertinent scientific and patent literature pertaining to colorant delivery in cheese and various types of colorant products on the market for cheese manufacture, and also considers interactions between colorant molecules and cheese components; various strategies for elimination of color transfer to whey during cheese manufacture are also discussed. © 2019 Institute of Food Technologists®

Link to article: <https://doi.org/10.1111/1541-4337.12519>

Funding Source: DPTC 2

Research Programme: RFA1

Polyhydroxyphenylvalerate/polycaprolactone nanofibers improve the life-span and mechanoresponse of human iPSC-derived cortical neuronal cells

Cerrone F.; Pozner T.; Siddiqui A.; Ceppi P.; Winner B.; Rajendiran M.; Babu R.; Ibrahim H.S.; Rodriguez B.J.; Winkler J.; Murphy K.J.; O'Connor K.E.

Materials Science and Engineering C, 2020

The physico-chemical characteristics of the extracellular matrix (ECM) cause mechanical cues that could elicit responses in the survival rate of cortical neuronal cells. Efficient neurite outgrowth in vitro, is critical for successful cultivation of cortical neuronal cells and the potential for attempts at regeneration of the central nervous system (CNS) in vivo. Relatively soft and hydrophilic, microbially synthesized aromatic polyester, polyhydroxyphenylvalerate (PHPV) was blended 50:50 with the stiff and hydrophobic polycaprolactone (PCL) and electrospun in microfibers for use in a 3D (CellCrown™) configuration and in a 2D coverslip coated configuration. This blend allows a 2.3-fold increase in the life-span of human induced pluripotent stem derived cortical neuronal cells (hiPS) compared to pure PCL fibers. HiPS-derived cortical neuronal cells grown on PHPV/PCL fibers show a 3.8-fold higher cumulative neurite elaboration compared to neurites grown on PCL fibers only. 96% of cortical neuronal cells die after 8 days of growth when plated on PCL fibers alone while >83% and 55% are alive on PHPV/PCL fibers on day 8 and day 17, respectively. An increased migration rate of cortical neuronal cells is also promoted by the blend compared to the PCL fibers alone. The critical survival rate improvement of hiPS derived cortical neuronal cells on PHPV/PCL blend holds promise in using these biocompatible nanofibers as implantable materials for regenerative purposes of an active cortical neuronal population after full maturation in vitro. © 2020 Elsevier B.V.

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Funding Source: DPTC 2

Research Programme: RFA1

Physicochemical and gelling properties of whey protein hydrolysates generated at 5 and 50 °C using Alcalase® and Neutrase®, effect of total solids and incubation time

Dermiki M.; FitzGerald R.J.

International Dairy Journal, 2020

Gelation temperature (T_g), apparent viscosity (η_{app}), turbidity and chromatography profiles of whey protein concentrate (WPC) and corresponding hydrolysates (WPHs) generated with Alcalase® and Neutrase® at different temperatures and total solids (TS) were compared. WPC incubated with Alcalase at 50 °C exhibited the highest rate and degree of hydrolysis (DH). After 4 h, the 5 °C Alcalase WPH (10% TS) had a DH and molecular mass distribution similar to those generated at 5 and 50 °C with Neutrase; however, the chromatography profiles differed. T_g was lower for WPHs than for WPC and depended on both the enzyme and TS. Moreover, the T_g s were lower for the Alcalase hydrolysates than for those generated with Neutrase. The η_{app} and turbidity properties of the hydrolysates depended on the enzyme used, solution TS and incubation temperature. Hydrolysates had lower η_{app} than WPC. The 50 °C Neutrase hydrolysates displayed lower turbidity than the corresponding Alcalase hydrolysates. © 2020 Elsevier Ltd

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Funding Source: DPTC 2

Research Programme: RFA1

Incorporating sustainable criteria in a dynamic multi-objective recommendation planning tool for a continuous manufacturing process: A dairy case study

Eccher C.; Geraghty J.

Journal of Manufacturing Systems, 2020

The activity of scheduling the production plan with the aim of achieving an optimal criterion has been explored in literature for several manufacturing sectors, in particular when it comes to solving scheduling NP-complete problems. In Dairy Manufacturing, determining an optimum criterion for the scheduling process has numerous internal and external challenges due to the complexity of this environment. The initial stages in the Dairy process are characterised by a continuous manufacturing environment and specific operational issues are observable: interruptions for the accomplishment of Cleaning-In-Place (CIP); a short raw material lifespan which demands a fast processing rate; and the stochastic raw material supply variation. By highlighting these three aspects, a critical trade-off emerges: CIP cycle-times heavily reduce the processing capacity, whereas the raw material processed requires an increase in available capacity due to the impact of seasonality, perishability and stochastic deliveries. Therefore, the scheduling plan must be dynamically readapted based on the current inventory, volume and frequency supplied, CIP cycle-times, maximum equipment running hours and downstream capacities. The aim of this research is to develop an integrated approach for generating equipment schedules under supply uncertainty typically observed in the dairy sector where criteria of sustainability are effortlessly incorporated for an improved decision-making process. An efficient Multi-objective Algorithm (MOA) combining conflicting key performance metrics such as minimising Work-In-Process (WIP), maximising Service Level Agreement (SLA), Utilisation and Energy consumption is proposed. The novelty consists of the ability to dynamically select trade-off criteria and visualise the optimum production plan according to the conditions defined by the decision-maker. The appropriate schedules are presented in a Pareto Frontier graph highlighting the entire non-dominance region according to the volume and frequency supplied. Even though sustainability metrics are usually ignored during production plan definitions, namely when a weak correlation between both environmental and profitable criteria is identified, the results demonstrate improved performance when both sustainable approaches are well explored. © 2020 The Society of Manufacturing Engineers

Link to article: <https://doi.org/10.1016/j.jmsy.2020.02.008>

Funding Source: DPTC 2

Research Programme: RFA1

Application of in silico approaches for the generation of milk protein-derived bioactive peptides

FitzGerald R.J.; Cermeño M.; Khalesi M.; Kleekayai T.; Amigo-Benavent M.

Journal of Functional Foods, 2020

Milk protein derived peptides have numerous well-documented bioactive properties. The conventional approach for the generation, identification and validation of bioactive peptides (BAPs) has involved (i) protein hydrolysis, (ii) bioactivity screening and (iii) validation in vivo. The low potency (in comparison to conventional drugs), susceptibility to breakdown during gastrointestinal transit and low intestinal permeability are key challenges in the development of highly bioactive food protein hydrolysates/peptides. However, the generation of potent and effective health enhancing hydrolysates/peptides can benefit from a range of in silico techniques including the application of structure bioactivity relationship modelling (e.g., quantitative structure activity relationship (QSAR) modelling), molecular docking and design of experiments (DOE) approaches to optimise BAP production and identification. Some examples of how these approaches have been employed in BAP discovery and generation will be outlined. © 2019 Elsevier Ltd

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Funding Source: DPTC 2

Research Programme: RFA1

In vitro characterisation of the antioxidative properties of whey protein hydrolysates generated under pH- And non pH-controlled conditions

Kleekayai T.; Le Gouic A.V.; Deracinois B.; Cudennec B.; FitzGerald R.J.

Foods, 2020

Bovine whey protein concentrate (WPC) was hydrolysed under pH-stat (ST) and non pH-controlled (free-fall, FF) conditions using Debitrase (DBT) and FlavorPro Whey (FPW). The resultant whey protein hydrolysates (WPHs) were assessed for the impact of hydrolysis conditions on the physicochemical and the in vitro antioxidant and intracellular reactive oxygen species (ROS) generation in oxidatively stressed HepG2 cells. Enzyme and hydrolysis condition dependent differences in the physicochemical properties of the hydrolysates were observed, however, the extent of hydrolysis was similar under ST and FF conditions. Significantly higher ($p < 0.05$) in vitro and cellular antioxidant activities were observed for the DBT compared to the FPW-WPHs. The WPHs generated under ST conditions displayed significantly higher ($p < 0.05$) oxygen radical absorbance capacity (ORAC) and Trolox equivalent antioxidant capacity (TEAC) values compared to the FF-WPHs. The impact of hydrolysis conditions was more pronounced in the in vitro compared to the cellular antioxidant assay. WPH peptide profiles (LC-MS/MS) were also enzyme and hydrolysis conditions dependent as illustrated in the case of β -lactoglobulin. Therefore, variation in the profiles of the peptides released may explain the observed differences in the antioxidant activity. Targeted generation of antioxidant hydrolysates needs to consider the hydrolysis conditions and the antioxidant assessment method employed. © 2020 by the authors.

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Funding Source: DPTC 2

Research Programme: RFA1

The effects of sequential heat treatment on microbial reduction and spore inactivation during milk processing

Li F.; Hunt K.; Buggy A.K.; Murphy K.M.; Ho Q.T.; O'Callaghan T.F.; Butler F.; Jordan K.; Tobin J.T.

International Dairy Journal, 2020

Sequential heating processes are commonly applied to milk by the dairy industry as part of their microbiological control strategy. Often pasteurisation at 72 °C is followed by a sequential high heat treatment step of up to 125 °C; however, such severe heat treatment can lead to reduced protein quality. Nine temperature combinations (80–90 °C) were evaluated to assess microbial reduction and whey protein nitrogen index values during pilot scale milk processing. A total of 110 bacterial isolates were identified to species level by 16S rDNA sequencing, with *Bacillus licheniformis* identified as the dominant species. While the experimental treatments did not achieve microbial reductions comparable with the control heating process, the results of this study provide a benchmark for milk processors relative to the effects of sequential heat treatments on milk and their impact on the survival of both thermally resistant microbial populations and thermally labile milk components during processing. © 2020 Elsevier Ltd

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Funding Source: DPTC 2

Research Programme: RFA1

Milk fat: opportunities, challenges and innovation

Mohan M.S.; O’Callaghan T.F.; Kelly P.; Hogan S.A.

Critical Reviews in Food Science and Nutrition, 2020

Milk fat is a high-value milk component that is processed mainly as butter, cheese, cream and whole milk powder. It is projected that approximately 35 million tonnes of milk fat will be produced globally by 2025. This surplus, enhances the need for diversification of milk fat products and the milk pool in general. Infant milk formula producers, for instance, have incorporated enzyme modified (“humanised”) milk fat and fat globule phospholipids to better mimic human milk fat structures. Minor components like mono- and di-glycerides from milk fat are increasingly utilized as emulsifiers, replacing palm esters in premium-priced food products. This review examines the chemistry of milk fat and the technologies employed for its modification, fractionation and enrichment. Emerging processing technologies such as ultrasound, high pressure processing, supercritical fluid extraction and fractionation, can be employed to improve the nutritional and functional attributes of milk fat. The potential of recent developments in biological intervention, through dietary manipulation of milk fatty acid profiles in cattle also offers significant promise. Finally, this review provides evidence to help redress the imbalance in reported associations between milk fat consumption and human health, and elucidates the health benefits associated with consumption of milk fat and dairy products. © 2020, © 2020 Taylor & Francis Group, LLC.

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Funding Source: DPTC 2

Research Programme: RFA1

Dynamic Mechanical Analysis as a Complementary Technique for Stickiness Determination in Model Whey Protein Powders

O'Donoghue, LT; Haque, MK; Hogan, SA; Laffir, FR; O'Mahony, JA; Murphy, EG

Foods, 2020

The α -relaxation temperatures (T_α), derived from the storage and loss moduli using dynamic mechanical analysis (DMA), were compared to methods for stickiness and glass transition determination for a selection of model whey protein concentrate (WPC) powders with varying protein contents. Glass transition temperatures (T_g) were determined using differential scanning calorimetry (DSC), and stickiness behavior was characterized using a fluidization technique. For the lower protein powders (WPC 20 and 35), the mechanical T_α determined from the storage modulus of the DMA (T_α onset) were in good agreement with the fluidization results, whereas for higher protein powders (WPC 50 and 65), the fluidization results compared better to the loss modulus results of the DMA (T_α peak). This study demonstrates that DMA has the potential to be a useful technique to complement stickiness characterization of dairy powders by providing an increased understanding of the mechanisms of stickiness.

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Funding Source: DPTC 2

Research Programme: RFA1

The effect of compositional changes due to seasonal variation on milk density and the determination of season-based density conversion factors for use in the dairy industry

Parmar P.; Lopez-Villalobos N.; Tobin J.T.; Murphy E.; McDonagh A.; Crowley S.V.; Kelly A.L.; Shalloo L.

Foods, 2020

The objective of this study was to determine the effect of seasonal variation on milk composition and establish an algorithm to predict density based on milk composition to enable the calculation of season-based density conversion calculations. A total of 1035 raw whole milk samples were collected from morning and evening milking of 60 spring-calving individual cows of different genetic groups, namely Jersey, Elite HF (Holstein Friesian) and National Average HF, once every two weeks for a period of 9 months (March November, 2018). The average mean and standard deviation for milk compositional traits were $4.72 \pm 1.30\%$ fat, $3.85 \pm 0.61\%$ protein and $4.69 \pm 0.30\%$ lactose and density was estimated at 1.0308 ± 0.002 g/cm³. The density of the milk samples was evaluated using three methods: a portable density meter, DMA 35; a standard desktop version, DMA 4500M; and an Association of Official Agricultural Chemists (AOAC) method using 100-mL glass pycnometers. Statistical analysis using a linear mixed model showed a significant difference in density of milk samples ($p < 0.05$) across seasonal and compositional variations adjusted for the effects of days in milk, parity, the feeding treatment, the genetic group and the measurement technique. The mean density values and standard error of mean estimated for milk samples in each season, i.e., spring, summer and autumn were 1.0304 ± 0.00008 g/cm³, 1.0314 ± 0.00005 g/cm³ and 1.0309 ± 0.00007 g/cm³, respectively. © 2020 by the authors. Licensee MDPI, Basel, Switzerland.

Link to article: <http://dx.doi.org/10.3390/foods9081004>

Funding Source: DPTC 2

Research Programme: RFA1

Application of a cascade membrane filtration process to standardise serum protein depleted cheese milk for cheddar cheese manufacture

Xia, XF; Tobin, JT; Sharma, P; Fenelon, M; McSweeney, PLH; Sheehan, JJ

International Dairy Journal, 2020

A cascade membrane filtration process including microfiltration (MF), ultrafiltration (UF) and reverse osmosis (RO) was used to fractionate skim milk into different streams. Significant quantities of lactose and minerals were removed to permeate after MF at 0.14 µm. Cheese milk, of similar casein content to the raw milk, was standardised simultaneously for casein, lactose, ash and total calcium from the membrane streams without requiring CaCl₂ and lactose addition. Serum protein depleted cheese milk of typical casein content had similar rennet coagulability, cheese composition, texture and yield to the control; milk of 1.5 × casein content had a faster coagulation rate and resulted in cheese of lower moisture content. On a dry matter basis, the serum protein content of MF permeate concentrated by UF was significantly higher than that in cheese whey (51.54% versus 5.63–9.45%), with significantly lower contents of ash (0.95% versus 7.11–7.53%) and lactose (9.50% versus 61.98–70.35%).

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Funding Source: DPTC 2

Research Programme: RFA1

Dilute phase pneumatic conveying of whey protein isolate powders: Particle breakage and its effects on bulk properties

Zhang F.; Olaleye A.K.; O'Mahony J.A.; Miao S.; Cronin K.

Advanced Powder Technology, 2020

Breakage of dairy powder during pneumatic conveying negatively affects the end-customer properties (scoop uniformity and reconstitution). A dilute phase pneumatic conveying system was built to conduct studies into this problem using whey protein isolate powder (WPI) as the test material. Effects of conveying air velocity (V), solid loading rate (SL), pipe bend radius (D), and initial particle size (d) on the level of attrition were experimentally studied. Four quality characteristics were measured before and after conveying: particle size distribution, tapped bulk density, flowability, and wettability. The damaged WPI agglomerates after conveying give rise to many porous holes exposed to the interstitial air. V is the most important input variable and breakage levels rise rapidly at higher airspeeds. The mean volume diameter $D[4,3]$ decreased by around 20% using the largest airspeed of 30 m/s. Powder breakage is also very sensitive to particle size. There appears to be a threshold size below which breakage is almost negligible. By contrast, SL and D show lesser influence on powder breakage. Reflecting the changes in particle size due to breakage, tapped bulk density increases whereas wettability decreases as a result of an increase in conveying air velocity. However, breakage does not show a significant effect on powder flowability as powder damage not only decreases particle size but also changes the particle's surface morphology. © 2020 The Society of Powder Technology Japan

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Funding Source: DPTC 2

Research Programme: RFA1

Characterization of norbixin and evaluation of its mobility through rennet-induced micellar casein concentrate gels as influenced by an electrical field

Alehosseini A.; Sharma P.; Kelly A.L.; Sheehan J.J.

Innovative Food Science and Emerging Technologies, 2021

In this study, application of an electrical field to facilitate the migration of norbixin molecules through rennet-induced micellar casein concentrate (MCC) and agar gels was evaluated. To determine the effect of selected parameters on norbixin penetration through the renneted casein gels, MCC, as a curd-like structure, was used to simplify the study of the process and evaluate the impact of each parameter on the migration of norbixin molecules separately. The physicochemical properties of norbixin solutions were characterized, followed by evaluation of the effects of changing gel composition on colorant mobility—influenced by an electrical field. Multi-angle dynamic light scattering enabled size measurement of norbixin solutions. A strong negative charge (-53.4 to -73.4 mV) was observed at all pH values examined. Localization of norbixin molecules within the aqueous phase was shown by confocal laser scanning microscopy. Norbixin solutions, characterized by LUMiSizer and light microscopy, showed the formation of aggregates (30 – 50 μm) in the presence of CaCl₂. Reducing pH and increasing calcium content considerably decreased norbixin penetration through the gels. The penetration rate of norbixin was considerably reduced ($\sim 60\%$) in the presence of 2% of calcium chloride. By reducing the pH from 6.58 to 5.37 and 6.60 to 5.30, the penetration rate of color through the gels containing 7.5 and 15% protein was reduced by ~ 90 and 80%, respectively. However, as salt concentrations increased (0% to 4%), the penetration rate of the colorant through MCC gels of 15% protein increased by 60%. Overall, this work shows that applying an electrical field is a promising approach which may considerably increase the migration rate of colorant through rennet-induced casein gels. © 2021

Link to article: <https://doi.org/10.1016/j.ifset.2021.102812>

Funding Source: DPTC 2

Research Programme: RFA1

Prevalence of intramammary antibiotic usage in dairy farming

Burke N.; Adley C.C.

Journal of Dairy Research, 2021

This research communication describes the lactating intramammary (IMM) antibiotic formulation most used by Irish dairy farmers at farm level through interviewing 202 dairy farmers. The IMM antibiotic usage data is not easily available to the researcher and farming community. This study determined that three commercial formulations (Synulox™, Tetra Delta™ and Terrexine) made up 81% of the products used at farm level. The formulation Synulox™ was the most used at 34% first preference and 32% second preference and contains amoxicillin/clavulanic, a standard broad spectrum antibiotic, for which mastitis pathogen resistance remains low. The aminoglycosides were used in four of the IMM formulations analysed, including Tetra Delta™ and Terrexine. Of the 12 antibiotics identified in the IMM formulations studied, three including cefalexin, benzylpenicillin and penethamate are classified as highly important antibiotics (HIA) by the World Health Organisation (WHO) whilst the other 8 (dihydrostreptomycin, streptomycin, neomycin, framycetin, kanamycin, amoxicillin/clavulanic acid, and cefquinome), are considered critically important (CIA) for use in human health. This study has generated knowledge of the preferences of lactating IMM formulations used at farm level. Copyright © The Author(s), 2021. Published by Cambridge University Press on behalf of Hannah Dairy Research Foundation.

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Funding Source: DPTC 2

Research Programme: RFA1

A comparison of analytical test methods in dairy processing

Burke N.; Zacharski K.; Adley C.C.; Southern M.

Food Control, 2021

Dairy quality strategies start at the beginning of a raw milk supply chain at farm level, but it is the obligation of the manufacturer at a dairy processing plant to ensure quality is upheld from intake to finished product. This is achieved by implementing robust quality systems, measured through sampling plans and analytical test methods. Influences on product quality and composition, and analytical test results within a dairy plant are multi-factorial including: seasonality; the quality of incoming milk and herd health; the level of skilled laboratory technicians; the level of production and the availability of equipment; and finally milk harvesting, transportation and handling. These factors, along with customer and regulatory requirements will determine the level and type of analytical testing required. In the dairy industry, manufacturers oftentimes pay little attention to the need for optimising analytical test strategies or improving laboratory operations, if it is not broken why fix it? The focus of this qualitative research was to differentiate the core current analytical test methods in use at three dairy manufacturing plants for the production of raw milk, skim milk and cream and skim milk powder (SMP). The main objective being to inform and educate each producer on best practice methods. Results displayed similarities across testing categories but demonstrated a range of traditional testing methods in the microbiological analysis compared to advanced instrumentation use in the chemical and compositional analytical category. The dairy industry needs to adapt to a modern, process focused quality system using industry 4.0 analytical processing regimes. © 2020 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

The influence of pre-heat treatment of skim milk on key quality attributes of fat filled milk powder made therefrom

Finnegan E.W.; Mahomud M.S.; Murphy E.G.; O'Mahony J.A.

International Journal of Dairy Technology, 2021

Key quality aspects of fat filled milk powders (FFMPs) produced with skim milk pre-heated at 93 °C × 60 s and 105 °C × 15 s (medium whey protein nitrogen index; WPNI), and 116 °C × 60 s and 136 °C × 15 s (high WPNI) were examined. The 136 °C × 15 s treatment produced a FFMP with threefold higher free fat, showed significantly ($P < 0.05$) lower solubility in coffee and exhibited extensive flecking on rehydration in water. Insoluble materials were observed using microscopy; flecks generated upon rehydration in water were composed of fat, whereas flecks/sediment in coffee were composed of undissolved powder particles. © 2021 Society of Dairy Technology

Link to article: <http://dx.doi.org/10.1111/1471-0307.12758>

Funding Source: DPTC 2

Research Programme: RFA1

Assessment of the response of indigenous microflora and inoculated *Bacillus licheniformis* endospores in reconstituted skim milk to microwave and conventional heating systems by flow cytometry

Li F.; Santillan-Urquiza E.; Cronin U.; O'Meara E.; McCarthy W.; Hogan S.A.; Wilkinson M.G.; Tobin J.T.

Journal of Dairy Science, 2021

Heat treatment is one of the most widely used processing technologies in the dairy industry. Its primary purpose is to destroy microorganisms, both pathogenic and spoilage, to ensure the product is safe and has a reasonable shelf life. In this study microwave volumetric heating (MVH) was compared with a conventional tubular heat exchanger (THE), in terms of the effects of each at a range of temperatures (75°C, 85°C, 95°C, 105°C, 115°C, and 125°C) on indigenous microflora viability and the germination of inoculated *Bacillus licheniformis* endospores in reconstituted skim milk. To assess the heat treatment-related effects on microbial viability, classical agar-based tests were applied to obtain the counts of 4 various microbiological groups including total bacterial, thermophilic bacterial, mesophilic aerobic bacterial endospore, and thermophilic aerobic bacterial endospore counts, and additional novel insights into cell permeability and spore germination profiles post-heat treatment were obtained using real-time flow cytometry (FC) methods. No significant differences in the plate counts of the indigenous microorganisms tested, the plate counts of the inoculated *B. licheniformis*, or the relative percentage of germinating endospores were observed between MVH- and THE-treated samples, at equal temperatures in the range specified above, indicating that both methods inactivated inoculated endospores to a similar degree (up to 70% as measured by FC and 5 log reduction as measured by plate counting for some treatments of inoculated endospores). Furthermore, increased cell permeability of indigenous microflora was observed by FC after MVH compared with THE treatment of uninoculated skim milk, which was reflected in lower total bacterial count at a treatment temperature of 105°C. This work demonstrates the utility of FC as a rapid method for assessing cell viability and spore inactivation for postthermal processing in dairy products and overall provides evidence that MVH is at least as effective at eliminating native microflora and inoculated *B. licheniformis* endospores as THE. © 2021 American Dairy Science Association

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Funding Source: DPTC 2

Research Programme: RFA1

Effect of temperature on raw whole milk density and its potential impact on milk payment in the dairy industry

Parmar P.; Lopez-Villalobos N.; Tobin J.T.; Murphy E.; Buckley F.; Crowley S.V.; Kelly A.L.; Shalloo L.

International Journal of Food Science and Technology, 2021

The objective of this study was to determine the effect of temperature on whole milk density measured at four different temperatures: 5, 10, 15, and 20 °C. A total of ninety-three individual milk samples were collected from morning milking of thirty-two Holstein Friesian dairy cows, of national average genetic merit, once every two weeks over a period of 4 weeks and were assessed by Fourier transform infrared spectroscopy for milk composition analysis. Density of the milk was evaluated using two different analytical methods: a portable density meter DMA35 and a standard desktop model DMA4500M (Anton Paar GmbH, UK). Milk density was analysed with a linear mixed model with the fixed effects of sampling period, temperature and analysis method; triple interaction of sampling period x analysis method x temperature; and the random effect of cow to account for repeated measures. The effect of temperature on milk density (ρ) was also evaluated including temperature (t) as covariate with linear and quadratic effects within each analytic method. The regression equation describing the curvature and density–temperature relationship for the DMA35 instrument was $\rho = 1.0338 - 0.00017T - 0.0000122T^2$ ($R^2 = 0.64$), while it was $\rho = 1.0334 + 0.000057T - 0.00001T^2$ ($R^2 = 0.61$) for DMA4500 instrument. The mean density determined with DMA4500 at 5 °C was 1.0334 g cm⁻³, with corresponding figures of 1.0330, 1.0320 and 1.0305 g cm⁻³ at 10, 15 and 20 °C, respectively. The milk density values obtained in this study at specific temperatures will help to address any bias in weight–volume calculations and thus may also improve the financial and operational control for the dairy processors in Ireland and internationally. © 2020 Institute of Food, Science and Technology (IFSTTF)

Link to article: <http://dx.doi.org/10.1111/ijfs.14869>

Funding Source: DPTC 2

Research Programme: RFA1

The effects of cow genetic group on the density of raw whole milk

Parmar P.; Lopez-Villalobos N.; Tobin J.T.; Murphy E.; Buckley F.; McDonagh A.;
O'Mahony J.A.; Crowley S.V.; Kelly A.L.; Shalloo L.

Irish Journal of Agricultural and Food Research, 2021

The density of milk is dependent upon various factors including temperature, processing conditions, and animal breed. This study evaluated the effect of different cow genetic groups, Jersey, elite Holstein Friesians (EHF), and national average Holstein Friesians (NAHF) on the compositional and physicochemical properties of milk. Approximately 1,040 representative (morning and evening) milk samples (~115 per month during 9 mo) were collected once every 2 wk. Milk composition was determined with a Bentley Dairyspec instrument. Data were analysed with a mixed linear model that included the fixed effects of sampling month, genetic group, interaction between month and genetic group and the random effects of cow to account for repeated measures on the same animal. Milk density was determined using three different analytical approaches – a portable and a standard desktop density meter and 100 cm³ calibrated glass pycnometers. Milk density was analysed with the same mixed model as for milk composition but including the analytical method as a fixed effect. Jersey cows had the greatest mean for fat content ($5.69 \pm 0.13\%$), followed by EHF ($4.81 \pm 0.16\%$) and NAHF ($4.30 \pm 0.15\%$). Milk density was significantly higher ($1.0313 \text{ g/cm}^3 \pm 0.00026$, $P < 0.05$) for the milk of Jersey breed when compared to the EHF ($1.0304 \pm 0.00026 \text{ g/cm}^3$) and NAHF ($1.0303 \pm 0.00024 \text{ g/cm}^3$) genetic groups. The results from this study can be used by farmers and dairy processors alike to enhance accuracy when calculating the quantity and value of milk solids depending upon the genetic merit of the animal/herd, and may also improve milk payment systems through relating milk solids content and density. © 2020 Authors. All rights reserved.

Link to article: <http://dx.doi.org/10.15212/ijafr-2020-0115>

Funding Source: DPTC 2

Research Programme: RFA1

Development and evaluation of a processing sector model for butter manufacture using a mass balance technique at two dairy processing sites

Parmar P.; Lopez-Villalobos N.; Tobin J.T.; Murphy E.; Crowley S.V.; Kelly A.L.; Shalloo L.

International Journal of Dairy Technology, 2021

The butter manufacturing process at two different commercial dairy processing sites in Ireland was evaluated using a mass balance approach to develop, evaluate and validate a processing sector model of the flow of milk fat from intake to final product. The mass balance was represented as a function of fat intake = fat in products + fat losses + recycled fat. Representative samples of all products, namely whole milk, cream, skim milk, butter, buttermilk and cleaning-in-place streams (cream silo flush, butter churn residue and sludge), were collected from two different sites. Milk fat levels and product quantities were measured to obtain the fat outputs. Total fat losses at the end of butter production ranged between 1.90% and 2.25% of the total fat input for both sites. Three different scenarios were examined to evaluate the model: S1 (Animal Breed) high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows were evaluated, for their effect on the net value of milk; S2 (Product Portfolio) a mixed product portfolio of cheese, butter and skim milk powder (SMP) was compared to a product portfolio comprised of butter alone; and S3 (Process Efficiency) the impact of varying process losses on net values of milk and the quantities of products produced was simulated. The value per 1000 L of milk for S1 was €410.69 and €393.20 for Elite and NA cow's milk, respectively. For S2, the butter-only product portfolio returned €355.10, whereas the mixed-products portfolio returned €369.60. Lastly, S3 corresponding returns for 1%, 2.2% and 5% losses was €365.90, €361.47 and €351.12, respectively. © 2020 Society of Dairy Technology

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Funding Source: DPTC 2

Research Programme: RFA1

Effect of thermal treatment on serum protein reduced micellar casein concentrate: An evaluation of rennet coagulability, cheese composition and yield

Xia, XF; Tobin, JT; Subhir, S; Fenelon, MA; McSweeney, PLH; Sheehan, JJ

International Dairy Journal, 2021

Microfiltration at 0.10 µm removed ~70.29% of serum proteins from milk and the resultant micellar casein concentrates (MCC) were subjected to no heat treatment (control), pasteurisation (72 °C × 15 s) and high heat treatment (HHT; 90 °C × 15s) before formulation of cheese milk for Cheddar cheese manufacture. MCC showed good heat stability due to low serum protein content. For cheese milk of typical casein content, both pasteurisation and HHT did not significantly influence pH, calcium distribution and rennet coagulability, or subsequent cheese composition and yield; although HHT elongated cheese make time significantly. On increasing casein content from 3.09% to 4.31%, there was no significant difference for rennet to cut time between cheeses made from milk with different thermal histories and casein contents. Overall, HHT of MCC had no significant impact on cheese make properties, cheese composition and yield of Cheddar cheese.

Link to article: <http://dx.doi.org/10.1016/j.idairyj.2020.104902>

Funding Source: DPTC 2

Research Programme: RFA1

ADSA Foundation Scholar Award: Materials science approach to the study of mechanical and diffusion properties in cheese

Sharma P.

Journal of Dairy Science, 2022

Dairy products can be manufactured in a variety of structural forms (e.g., liquid, semi-solids, and solids). Although liquid milk is a colloidal dispersion of fat and protein in the serum portion, it can easily be converted into a soft gel (yogurt) upon acid coagulation. Similarly, cheese, a rennet-coagulated, casein-rich fraction of milk, falls in the category of semi-solid foods. Structurally, all of these materials are complex in nature because of interactions between protein, fat, and water components. The structural origin of these diversified food textures is derived from the way that various food constituents are arranged to form a unique body or mass. Food materials science helps in the assessment of structural arrangements of these molecules at various length scales. This article focuses on the use of various materials science approaches for understanding the fundamental relationship between process, structure, and property in solving critical issues that pertain to the dairy industry and academia. © 2022 American Dairy Science Association

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Funding Source: DPTC 2

Research Programme: RFA1

State of the art and prospects of zeolites and metal organic frameworks (MOFs) for nitrogen and phosphorus removal in dairy wastewater

Bouanga Boudiombo J.S.; Madden D.G.; Cusack B.; Cronin P.; Ryan A.

Chemosphere, 2023

Water is an essential resource for humans, animals, and plants. Water is also necessary for the manufacture of many products such as milk, textiles, paper, and pharmaceutical composites. During manufacturing, some industries generate a large amount of wastewater containing numerous contaminants. In the dairy industry, for each litre of drinking milk produced, about 10 L of wastewater is generated. Despite this environmental footprint, the production of milk, butter, ice cream, baby formula, etc., are essential in many households. Common contaminants in dairy wastewater include high biological oxygen demand (BOD), chemical oxygen demand (COD), salts as well as nitrogen and phosphorus derivatives. Nitrogen and phosphorus discharges are one of the leading causes in the eutrophication of rivers and oceans. Porous materials have long held significant potential as a disruptive technology for wastewater treatment. However, thus far they have been understudied for use in dairy wastewater treatment. Ordered porous materials, such as zeolites and metal organic frameworks (MOFs), represent classes of porous materials with significant potential for the removal of nitrogen and phosphorus. This review explores the different zeolites and MOFs applied in the removal of nitrogen and phosphorus from wastewater and the prospect of their potential for use in wastewater management in the dairy industry. © 2023 Elsevier Ltd

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Funding Source: DPTC 2

Research Programme: RFA1

Influence of processing temperature on plasmin activity and proteolysis in process streams from cold microfiltration of skim milk

France T.C.; Kelly A.L.; Crowley S.V.; O'Mahony J.A.

International Dairy Journal, 2023

Plasmin in process streams derived from microfiltration of skim milk can result in casein hydrolysis, potentially affecting the quality and functionality of ingredients produced. To determine whether partitioning of plasmin into permeates was impacted by processing temperature, cold microfiltration of skim milk was investigated at 4, 8 and 12 °C. Permeate generated at 4 °C had the highest plasmin activity (0.0185 AMC units mL⁻¹). When plasmin activity was expressed relative to β-casein content, differences in the extent of dissociation of β-casein, at the different processing temperatures, did not influence plasmin activity in the resulting permeate streams. Throughout storage at 37 °C, all retentates exhibited extensive plasmin-mediated hydrolysis of β- and αS2-casein; αS1-casein to a lesser extent. Particle size measurements before and after plasmin-mediated hydrolysis of permeates indicated that the proteolysis products of β-casein, γ-caseins, retained the ability to self-associate on heating, with particle size increasing with increasing temperature. © 2023

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Funding Source: DPTC 2

Research Programme: RFA1

Technological solutions and adaptive processing tools to mitigate the impact of seasonal variations in milk composition on Cheddar cheese production—A review

Galli B.D.; Hamed A.M.; Sheehan J.J.; King N.; Abdel-Hamid M.; Romeih E.

International Journal of Dairy Technology, 2023

Variability in milk composition and physicochemical properties impact both manufacturing process performance and the end-use functionality of cheese. Such variability may be attributed to seasonal calving patterns, production systems, breed, weather patterns, and feed type and significantly impacts concentrations of macro- and microconstituents and ultimately cheese composition and quality. This article reviews technological approaches (e.g., milk standardisation protocols and calcium addition), and predictive strategies (e.g., predictive models for coagulation and curd cutting time, in-line sensors), used to mitigate the effects of seasonal changes in milk composition and their impact on process efficacy and functionality in Cheddar cheese manufacturing. © 2023 The Authors. International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Funding Source: DPTC 2

Research Programme: RFA1

Draft Genome Sequences of *Bacillus licheniformis* and *Bacillus paralicheniformis* Strains Isolated from Irish Skim Milk Powder

Lourenco A.; Li F.; Quijada N.M.; Duffy G.; Tobin J.T.; Butler F.; Jordan K.; O'Brien T.

Microbiology Resource Announcements, 2023

Nineteen *Bacillus licheniformis* strains and four strains of the closely related species *Bacillus paralicheniformis* were isolated from a variety of Irish medium-heat skim milk powders. The draft genome sequences of these 23 isolates provide valuable genetic data for research work relevant to dairy products and process development. The isolates are available at Teagasc. © Crown copyright 2023.

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Funding Source: DPTC 2

Research Programme: RFA1

A porous-crust drying model for a single dairy droplet

O'Connell K.; Olaleye A.K.; Van den Akker H.E.A.

Chemical Engineering Research and Design, 2023

The development of a novel numerical model for droplet drying is the topic of this paper. The three main stages of droplet drying are distinguished, viz. unhindered evaporation of a 'wet' particle (the droplet), restricted drying at a falling rate due to the formation of a crust around a wet core, and inert heating of the dry porous particle. Each stage is mathematically detailed to replicate all phenomena occurring throughout the drying process. The focus, however, is on the falling rate drying regime which is described in terms of Stefan diffusion of water vapour through the pores of a thickening crust. To this end, the model needs the material properties. This permits the droplet characteristics to be determined by composition rather than through single-droplet drying experiments. Finally, the model is validated against five of such experiments from literature using skim milk. Good agreement is found at each comparative case for the particle mass and temperature throughout the various drying regimes providing that for good reasons in three cases a lower drying air temperature is applied than reported for the experiments. The model is capable of predicting the entire drying process at low computational cost and without requiring empirical input. © 2023 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

From lab-based to in-line: Analytical tools for the characterization of whey protein denaturation and aggregation—A review

Finnegan E.W.; Goulding D.A.; O'Callaghan T.F.; O'Mahony J.A.

Comprehensive Reviews in Food Science and Food Safety, 2024

Whey protein denaturation and aggregation have long been areas of research interest to the dairy industry, having significant implications for process performance and final product functionality and quality. As such, a significant number of analytical techniques have been developed or adapted to assess and characterize levels of whey protein denaturation and aggregation, to either maximize processing efficiency or create products with enhanced functionality (both technological and biological). This review aims to collate and critique these approaches based on their analytical principles and outline their application for the assessment of denaturation and aggregation. This review also provides insights into recent developments in process analytical technologies relating to whey protein denaturation and aggregation, whereby some of the analytical methods have been adapted to enable measurements in-line. Developments in this area will enable more live, in-process data to be generated, which will subsequently allow more adaptive processing, enabling improved product quality and processing efficiency. Along with the applicability of these techniques for the assessment of whey protein denaturation and aggregation, limitations are also presented to help assess the suitability of each analytical technique for specific areas of interest. © 2024 The Authors. *Comprehensive Reviews in Food Science and Food Safety* published by Wiley Periodicals LLC on behalf of Institute of Food Technologists.

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Funding Source: DPTC 2

Research Programme: RFA1

Physicochemical properties of micellar casein retentates generated at different microfiltration temperatures

France T.C.; Bot F.; Kelly A.L.; Crowley S.V.; O'Mahony J.A.

Journal of Dairy Science, 2024

Processing temperature has a significant influence on the composition and functionality of the resulting streams following microfiltration (MF) of skim milk. In this study, MF and diafiltration (DF) were performed at 4 or 50°C to produce β -casein (β -CN)-depleted and nondepleted (i.e., native casein profile) micellar casein isolate retentates, respectively. Microfiltration combined with extensive DF resulted in a 40% depletion of β -CN at 4°C, whereas no β -CN depletion occurred at 50°C. Microfiltration at 4°C led to higher transmission of calcium into permeates, with retentate generated at 4°C containing less total calcium compared with retentate generated at 50°C, based on the volume of retentate remaining. Higher heat stability at 120°C was measured for retentates generated at 4°C compared with those at 50°C, across all pH values measured. Retentates generated at 4°C also had significantly lower ionic calcium values at each pH compared with those generated at 50°C. Higher apparent viscosities at 4°C were measured for retentates generated at 4°C compared with retentates generated at 50°C, likely due to increased voluminosity of β -CN-depleted casein micelles. The results of this study provide new information on how changing the composition of MF retentate, by appropriate control of processing temperature and DF, can alter physicochemical properties of casein micelles, with potential implications for ingredient functionality. © 2024 American Dairy Science Association

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Funding Source: DPTC 2

Research Programme: RFA1

Adaptive and predictive approaches to mitigate the impact of milk seasonality on composition, processing technologies and quality of milk powders

Hamed A.M.; Galli B.; Hogan S.A.; Abdel-Hamid M.; Romeih E.

International Journal of Dairy Technology, 2024

The diverse composition of milk throughout the year can potentially impact subsequent processing in the dairy industry and the quality of the final products. This fluctuation in milk composition is influenced by direct and indirect factors like seasonal variability. Milk powders are produced through a complex process and are intended for various end uses and require consistency of composition along with desirable physical and functional properties. This review aims to report technical challenges and limitations related to the functional and quality characteristics of milk powder and highlight recent approaches used to mitigate seasonally related differences in milk composition. © 2024 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Funding Source: DPTC 2

Research Programme: RFA1

Examination of the impact of using lactose or permeate for protein standardisation of skimmed milk on viscosity characteristics during evaporation

Long T.; Finnegan E.W.; Girivasan A.; Tarapata J.; McCarthy N.A.; O'Mahony J.A.; O'Callaghan T.F.

International Journal of Dairy Technology, 2024

The effect of permeate and lactose for protein standardisation of skim milk concentrate, was investigated during evaporation. Analysis of the viscosity–total solids (TS) profiles of each treatment demonstrated that the unstandardised protein sample had the greatest increase in viscosity, followed by permeate and finally lactose-standardised samples. Heat stability and acid buffering capacity of the permeate standardised skim milk samples were higher when compared to the lactose-standardised samples. This work identifies how standardisation media can impact the viscosity of concentrated skim milk, enabling higher TS at the evaporator outlet in some circumstances which could have the potential to improve overall process efficiency. Furthermore this work demonstrates that, evaporating to a target viscosity rather than target TS would offer greater control and consistency during processing. © 2024 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Research Programme: RFA1

Prediction of milk composition using multivariate chemometric modelling of infrared, Raman, and fluorescence spectroscopic data: A review

Mohammadi S.; Gowen A.; Luo J.; O'Donnell C.

Food Control, 2024

Quality assessment of milk which is a comprehensive source of nutrients for humans and an important raw material for other dairy products is required in the dairy industry. Rapid, cost-effective, and non-destructive spectroscopic techniques are more preferable than classic wet chemistry approaches for milk analysis. The objective of this work was to review the prediction of milk composition including macronutrients such as fat, protein and lactose and micronutrients such as fatty acids and vitamins using multivariate chemometric modelling of Near Infrared (NIR), Mid Infrared (MIR), fluorescence, and Raman spectral data and data fusion approaches. Literature sources describing spectroscopic analysis of milk samples and the application of multivariate data analysis methods are outlined in this literature review. In addition, the importance of data fusion strategies employed for combining different spectroscopic techniques are reviewed to evaluate their potential to improve the accuracy of the prediction models developed. Recent research studies have demonstrated that the use of data fusion strategies improves the performance of milk composition prediction models developed. © 2024 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

Vibrational spectroscopy data fusion for enhanced classification of different milk types

Mohammadi S.; Gowen A.; O'Donnell C.

Heliyon, 2024

The aim of this study is to classify seven types of Irish milk (butter, fresh, heart active, lactose free, light, protein, and slimline), supplied by a specific company, using vibrational spectroscopy methods: Near infrared (NIR), mid infrared (MIR), and Raman spectroscopy. In this regard, chemometric methods were used, and the impact of spectral data fusion on prediction accuracy was evaluated. A total of 105 samples were tested, with 21 used in the test set. The study assessed principal component analysis (PCA), partial least square discriminant analysis (PLS-DA), and sequential and orthogonalized partial least squares linear discriminant analysis (SO-PLS-LDA) for classifying different milk types. The prediction accuracy, when applying PLS-DA on individual blocks of data and low-level fused data, did not exceed 85.71 %. However, implementing the SO-PLS-LDA strategy significantly improved the accuracy to 95 %, suggesting a promising method for the development of classification models for milk using data fusion strategies. © 2024 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

The Impact of pH on Fouling and Related Physicochemical Properties of Skim Milk Concentrate during Heat Treatment Using a Laboratory-Scale Fouling Rig

Murphy T.R.; Finnegan E.W.; Tarapata J.; O'Callaghan T.F.; O'Mahony J.A.

Foods, 2024

The objective of this study was to investigate the effect of pH (6.1, 6.3, 6.5, and 6.7) on heat-induced changes in concentrated skim milk as related to fouling in heat exchangers. Skim milk (30%, w/w, total solids) was recirculated in a laboratory-scale fouling rig at an initial target temperature of 85 °C for 90 min to simulate thermal processing and preheating of evaporated liquid concentrate feeds in dairy processing. This study investigated key changes in relevant physicochemical properties, such as viscosity, particle size, and sedimentation, as major contributors to fouling at lower pHs (6.1 and 6.3). Additionally, protein aggregation and calcium phosphate precipitation were identified as significant contributors to fouling deposits. Possible strategies to mitigate fouling were determined, including optimizing pH and adjusting heat treatment parameters to minimize protein denaturation and mineral deposition. The findings indicate that carefully controlling pH and processing parameters can greatly enhance the efficiency of milk concentration by evaporation and tailor finished product quality. Moreover, this study showed that monitoring of CIP solutions for protein content and turbidity provides valuable information on the intensity of fouling and the efficiency of cleaning. © 2024 by the authors.

Link to article: <https://doi.org/10.3390/foods13193100>

Funding Source: DPTC 2

Research Programme: RFA1

A Review on MIR, NIR, Fluorescence and Raman Spectroscopy Combined with Chemometric Modeling to Predict the Functional Properties of Raw Bovine Milk

Ní Fhuaráin A.M.; O'Donnell C.P.; Luo J.; Gowen A.A.

ACS Food Science and Technology, 2024

Spectroscopic methods, such as Mid-Infrared (MIR), Near-Infrared (NIR), fluorescence and Raman spectroscopy are rapid, inexpensive and nondestructive. Traditionally, mainly MIR and NIR spectroscopy have been employed to predict the compositional properties of milk. However, measurement of the key functional properties of milk is of high industry relevance. In this review, studies on the use of spectroscopic techniques for predicting milk functional properties are compared and reported models are outlined. The challenges of employing spectroscopy in functionality applications are discussed. For pH and curd yield, some of the MIR models display a robust prediction performance. With further model validation, calibrations for these properties could potentially be added to existing MIR instruments in the industry. Despite fluorescence and NIR spectroscopy being used for many dairy applications, their use for milk functionality is limited currently. As Raman spectroscopy is sensitive to the components of raw milk, it has potential for predicting milk functional properties. © 2024 The Authors. Published by American Chemical Society.

Link to article: <https://doi.org/10.1021/acsfoodscitech.4c00130>

Funding Source: DPTC 2

Research Programme: RFA1

Transglutaminase in Dairy Processing

Romeih E.; Kieliszek M.; Hebishy E.

Transglutaminase: Fundamentals and Applications, 2024

Transglutaminases derived from microbial sources are collectively known as microbial transglutaminases (MTGases). MTGases are widely known to modify food proteins, through the formation of both inter- and intramolecular isopeptide bonds, yielding a modified protein structures with altered functional properties in food systems. These modifications can improve protein functionality (gelation, solubility, foaming, emulsification, water binding, viscosity, and thermal stability) and nutritional characteristics (allergenicity and antioxidant activity). A comprehensive overview of the potential applications of MTGase in dairy products including fermented milks, cheese products, ice cream, milk powders, and other dairy-based products as well as relevant patents is discussed in this chapter. The frontiers of knowledge and technology in this chapter not only contribute substantially to establishing a foundation to develop an innovative dairy product and/or ingredient that possesses bio- and multifunctional properties but also offer promising options for improving dairy processing. © 2024 Elsevier Inc. All rights reserved.

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Funding Source: DPTC 2

Research Programme: RFA1

Efficient mass-preserving finite volume approach for the rennet-induced coagulation equation

Singh M.; Sriwastav N.; Shardt O.

Chaos, Solitons and Fractals, 2024

The coagulation of casein micelles caused by enzymes is a critical step in the dairy industry for cheese manufacture. During enzymatic coagulation of milk, three processes occur: enzymic proteolysis, coagulation, and gelation. This study presents the first numerical approach based on a finite volume scheme for describing the enzyme-induced coagulation of casein micelles. The finite volume scheme is mainly concerned with ensuring mass conservation and developed on the assumption that the particles are concentrated on the mean of each cell of the discretization. The key advantages of the new technique are its simple mathematical formulation and its robustness that allow it to be implemented on any type of grid and tailored to different coagulation kernels. The accuracy of the new approach is compared with newly derived analytical results for several gelling and non-gelling coagulation kernels. The comparison demonstrates that the new approach closely matches the exact results. In order to analyse the convergence behaviour of different order moments, various refined non-uniform grids have been taken into consideration. © 2024 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

Physicochemical properties and stability of milk permeate as influenced by ultrafiltration processing parameters

Tsermoula P.; Barone G.; O'Mahony J.A.

International Dairy Journal, 2024

The objective of this study was to determine the effect of membrane molecular weight cut-off (5 and 10 kDa), and filtration temperature (25, 15 and 10 °C) on the physicochemical properties of milk permeate (MP). Although MPs produced had a similar gross chemical composition, MP produced with the 10 kDa membrane at 25 °C had the lowest pH (6.64) and ionic calcium content (2.33 mM), while MP produced with the 5 kDa membrane at 10 °C had the highest levels of 6.82 and 2.85 mM for pH and ionic calcium, respectively. Incubation of MPs at 60 °C resulted in precipitation, with MPs produced with the 10 kDa membrane having larger, less soluble and thermodynamically more stable particles than MPs produced with the 5 kDa membrane. These results demonstrate that filtration parameters significantly affected the physicochemical properties of MPs, with implications for downstream processing. © 2024 The Authors

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Funding Source: DPTC 2

Research Programme: RFA1

Seasonal and geographical impact on the Irish raw milk microbiota correlates with chemical composition and climatic variables

Yap M.; O'Sullivan O.; O'Toole P.W.; Sheehan J.J.; Fenelon M.A.; Cotter P.D.

mSystems, 2024

Season and location have previously been shown to be associated with differences in the microbiota of raw milk, especially in milk from pasture-based systems. Here, we further advance research in this area by examining differences in the raw milk microbiota from several locations across Ireland over 12 months, and by investigating microbiota associations with climatic variables and chemical composition. Shotgun metagenomic sequencing was used to investigate the microbiota of raw milk collected from nine locations (n = 241). Concurrent chemical analysis of the protein, fat, lactose, total solids, nonprotein nitrogen contents, and titratable acidity (TA) of the same raw milk were performed. Although the raw milk microbiota was highly diverse, a core microbiota was found, with *Pseudomonas*_E, *Lactococcus*, *Acinetobacter*, and *Leuconostoc* present in all samples. Microbiota diversity significantly differed by season and location, with differences in seasonality and geography corresponding to 11.8% and 10.5% of the variation in the microbiota. Functional and antibiotic resistance profiles also varied across season and location. The analysis of other metadata revealed additional interactions, such as an association between mean daily air and grass temperatures with the abundance of spoilage taxa like *Pseudomonas* species. Correlations were identified between pathogenic, mastitis-related species, fat content, and the number of sun hours, suggesting a seasonal effect. Ultimately, this study expands our understanding of the interconnected nature of the microbiota, environment/climate variables, and chemical composition of raw milk and provides evidence of a season- and location-specific microbiota. **IMPORTANCE** The microbiota of raw milk is influenced by many factors that encourage or prevent the introduction and growth of both beneficial and undesirable microorganisms. The seasonal and geographical impacts on the microbial communities of raw milk have been previously seen, but the relationships with environmental factors and the chemical composition has yet to be investigated. In this year-long study, we found that while raw milk is highly diverse, a core microbiota was detected for Irish raw milk, with strong evidence of seasonal and geographical influence. We also found associations between groups of microorganisms, environmental factors, and milk composition, which expand current knowledge on the relationships between microbial and chemical composition and the climate. These results provide evidence for the development of a tool to allow for the prediction of raw milk quality and safety. © 2024 American Society for Microbiology. All rights reserved.

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Funding Source: DPTC 2

Research Programme: RFA1

Influence of hybrid lactose-permeate standardisation on the physicochemical and functional properties of skim milk and concentrates

Cippollini R.; Pecchini M.; Bot F.; O'Mahony J.A.

International Dairy Journal, 2025

Standardisation of protein content is important in ensuring consistent composition and functionality of dairy powders, especially those made from a seasonal milk supply. This study investigated the effects of hybrid standardisation media, composed of lactose and permeate mixtures, on the physicochemical and functional properties of skim milk concentrates. The protein content target was 3.5 %, using five different permeate:lactose (P:L) ratios: 100P, 75P:25L, 50P:50L, 25P:75L, and 100L (at 8 % total solids (TS)), followed by evaporation to 52 % TS. Results showed that higher P:L ratios led to higher viscosity of skim milk concentrates and enhanced the heat stability of protein-standardised milk at pH in the ranges 6.4–6.7 and 7.0–7.2. Mineral analysis reflected higher concentrations of calcium, phosphorous, and trace minerals for higher P:L ratios, contributing to higher buffering capacity, increased micellar hydration and larger particle size post-evaporation. These findings demonstrate the potential of hybrid standardisation media to optimise processing conditions in milk powder production, balancing enhanced heat stability and viscosity development of milk concentrates. © 2025

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Funding Source: DPTC 2

Research Programme: RFA1

A meshfree approach for the rennet-induced coagulation equation: Spline based multistage Bernstein collocation method and its convergence analysis

Sriwastav N.; Das A.; Shardt O.; Kumar J.; Singh M.

Applied Mathematical Modelling, 2025

The initial phases of milk coagulation for cheese manufacturing can be tracked by an integro-differential equation known as a population balance equation. In this article, a new analytical approach using multistage Bernstein polynomials is presented to solve a rennet-induced coagulation equation for the first time. The existence of the solution and convergence analysis of the proposed approach are discussed in detail to support the mathematical formulation. Our main interest is in computing the integral moments, such as the number and total volume/mass of casein micelles over time. These moments are evaluated by approximating them with the linear combinations of Bernstein polynomials that involve unknown coefficients. Furthermore, the unknown coefficients are determined by selecting an appropriate number of collocation points, based on the considered time span of the process. To test the accuracy and efficiency of the new approach, the new analytical solutions for the integral moments are obtained for constant, sum and product coagulation kernels and results are verified by comparing with the existing finite volume scheme and Picard's method. © 2025

Link to article: <https://doi.org/10.1016/j.apm.2025.116035>

Funding Source: DPTC 2

Research Programme: RFA1

Approaches for Measuring and Predicting Fouling During Thermal Processing of Dairy Solutions

Tarapata J.; Murphy T.R.; Finnegan E.W.; O'Callaghan T.F.; O'Mahony J.A.

Comprehensive Reviews in Food Science and Food Safety, 2025

Fouling during the thermal processing of dairy products remains a significant challenge, reducing operational efficiency, increasing energy consumption, and complicating cleaning cycles. This review critically assesses current methods for measuring and predicting fouling during thermal processing in the dairy industry, emphasizing scientific principles, technical maturity, and industrial applicability. Unlike existing reviews, which are mostly focused on fouling quantification, this work highlights the shift toward prediction-driven approaches for fouling control and minimization. Traditional measurement techniques, such as monitoring thermal resistance and pressure drop, are evaluated alongside emerging methods, including acoustic, spectroscopic, and electrochemical sensors. Their respective limitations and strengths are discussed in terms of sensitivity, scalability, and industrial robustness. Advanced predictive tools, including deep learning, computational fluid dynamics, and dimensional analysis techniques, are explored for their ability to model the dynamic nature of fouling and support real-time decision-making. The integration of artificial intelligence with real-time process data acquisition is identified as a key innovation for improving fouling management and optimizing cleaning schedules. The review also considers the importance of small-scale experimental systems in linking laboratory-scale research with industrial applications. Development and utilization of tools for enhanced process efficiency through prediction, prevention, and control of in-process fouling are growing. Greater control in this regard offers substantial opportunity to meet future challenges in process optimization, shorten cleaning-in-place times, and advance sustainable dairy manufacturing through real-time monitoring, predictive analytics, and industrial-scale implementation. Addressing these challenges will require a multidisciplinary approach between researchers, engineers, and industry stakeholders to translate emerging technologies into practical, scalable solutions. © 2025 The Author(s). Comprehensive Reviews in Food Science and Food Safety published by Wiley Periodicals LLC on behalf of Institute of Food Technologists.

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Funding Source: DPTC 2

Research Programme: RFA1

Flecking of fat-filled milk powders

Tarapata J.; Timlin M.; McCarthy N.A.; O'Mahony J.A.

Comprehensive Reviews in Food Science and Food Safety, 2025

Flecking is an insolubility issue in fat-containing milk powders. The undissolved particles (flecks) are of different shapes and structures, primarily composed of fat and/or protein. The occurrence of flecking in reconstituted milk powders negatively impacts the visual appearance and overall quality of the final product, thereby influencing consumer acceptance and brand trust. Standard quality control measures, like wettability or insolubility, and analysis including rehydration testing are important but not sufficient in predicting, identifying and/or quantifying flecking, often necessitating additional measures to be implemented. Suitable additional analyses for flecking include confocal laser scanning microscopy, electron microscopy, particle size, and density analysis. However, it is crucial to highlight that merely tightening quality control parameters is insufficient to combat flecking. This approach does not allow for the implementation of rapid solutions when the issue is detected at the final stages of quality assessment. To effectively address fleck formation, it is necessary to scrutinize unit operations and identify precisely where, and how, in the process flecks are formed. The issue often requires reformulation and/or engineering interventions, making the final product more robust and resilient to fleck formation. To date, protein denaturation/aggregation and emulsion instability are proposed as major mechanisms governing fleck formation. Additionally, the effect of seasonality of milk chemical composition and reconstitution medium (water/coffee/tea) are other important factors. This work aims to review flecking in reconstituted fat-filled milk powder solutions by interrogating the production process, including the skim milk base wet and dry processing, alongside the powder storage conditions and reconstitution methods, and thereby identify strategies for the control of flecking. © 2025 Institute of Food Technologists®.

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Funding Source: DPTC 2

Research Programme: RFA1

Adaptive and predictive approaches to mitigate the impact of milk seasonality on composition, processing technologies and quality of milk powders

Hamed A.M.; Galli B.; Hogan S.A.; Abdel-Hamid M.; Romeih E.

International Journal of Dairy Technology, 2025

The diverse composition of milk throughout the year can potentially impact subsequent processing in the dairy industry and the quality of the final products. This fluctuation in milk composition is influenced by direct and indirect factors like seasonal variability. Milk powders are produced through a complex process and are intended for various end uses and require consistency of composition along with desirable physical and functional properties. This review aims to report technical challenges and limitations related to the functional and quality characteristics of milk powder and highlight recent approaches used to mitigate seasonally related differences in milk composition. © 2024 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Research Programme: RFA1

Techniques to Detect and Quantify the Bacterial Metalloprotease AprX in Bovine Milk: A Review

Sinha A.; Kelly A.L.

Comprehensive Reviews in Food Science and Food Safety, 2026

The heat-stable metalloprotease AprX, secreted by psychrotrophic *Pseudomonas* spp., is a major cause of quality deterioration in dairy products, particularly ultrahigh temperature (UHT) milk. This review synthesizes the evolution and current state of detection and quantification techniques for AprX in bovine milk, covering traditional immunological assays, enzymatic activity measurements, and zymography, alongside modern molecular approaches such as PCR-based methods, mass spectrometry, and advanced biosensors. Highly sensitive systems, including indirect ELISA (LOD 21.0 ng/mL), multiplex qPCR, and biosensor platforms, have enhanced the ability to detect AprX activity at low concentrations. Comparative analysis evaluates these methods in terms of sensitivity, specificity, turnaround time, cost, ease of use, and industrial applicability. Emerging directions such as multiomics integration, biosensor miniaturization, and artificial intelligence-driven data interpretation are also discussed. By critically assessing available and emerging tools, this review supports dairy scientists and industry stakeholders in selecting optimal strategies to detect, monitor, and mitigate AprX-associated spoilage in milk and dairy products. © 2026 The Author(s). Comprehensive Reviews in Food Science and Food Safety published by Wiley Periodicals LLC on behalf of Institute of Food Technologists.

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Funding Source: DPTC 2

Research Programme: RFA1

Examination of the impact of using lactose or permeate for protein standardisation of skimmed milk on viscosity characteristics during evaporation

Long T.; Finnegan E.W.; Girivasan A.; Tarapata J.; McCarthy N.A.; O'Mahony J.A.; O'Callaghan T.F.

International Journal of Dairy Technology, 2026

The effect of permeate and lactose for protein standardisation of skim milk concentrate, was investigated during evaporation. Analysis of the viscosity–total solids (TS) profiles of each treatment demonstrated that the unstandardised protein sample had the greatest increase in viscosity, followed by permeate and finally lactose-standardised samples. Heat stability and acid buffering capacity of the permeate standardised skim milk samples were higher when compared to the lactose-standardised samples. This work identifies how standardisation media can impact the viscosity of concentrated skim milk, enabling higher TS at the evaporator outlet in some circumstances which could have the potential to improve overall process efficiency. Furthermore this work demonstrates that, evaporating to a target viscosity rather than target TS would offer greater control and consistency during processing. © 2024 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Funding Source: DPTC 2

Research Programme: RFA1

Seasonal variation in Irish industry bovine milk composition characterised by chemical analysis and near-infrared spectroscopy: A 12-month study

Zhang K.; O'Donnell C.P.; McGrath A.; Sheehan J.J.; O'Shea N.

International Journal of Dairy Technology, 2026

Rationale: Milk composition is an important factor affecting final cheese quality and yield, plant efficiencies, as well as determining the overall profitability of cheese processing plants. However, milk composition varies by season. **Aim:** This study will investigate seasonal variation in key compositional components in Irish bovine milk and evaluate the potential of near-infrared (NIR) spectroscopy for the rapid prediction of raw milk composition for informing upstream processing. **Methods:** This study presents a comprehensive analysis of seasonal fluctuations in key milk components by examining 555 homogenised bovine milk samples from industry-scale silos across Ireland over a 12-month period. Simultaneously, the feasibility of near-infrared (NIR) spectroscopy for rapid detection of key compositional components (i.e. total solids, total protein, fat, casein, lactose and ionic calcium) in raw milk prior to chymosin-induced coagulation was evaluated. Principal component analysis (PCA) and partial least squares (PLS) regression were developed, with PLSR being assessed using coefficient of determination (R^2_{cv}), root mean square error of cross-validation ($RMSE_{cv}$), ratio of standard error of prediction to sample standard deviation (RPD) and range of error ratio. **Major Findings:** Wet chemistry analysis revealed clear seasonal trends in milk composition. PCA visualised distinct seasonal clustering, with winter samples clearly separated from spring and summer. High predictive accuracy was achieved for fat content ($R^2_{cv} = 0.92$, $RMSE_{cv} = 0.11$, $RER_{cv} = 14.53$, $RPD_{cv} = 3.46$), with good performance also observed for total protein and casein content prediction ($R^2_{cv} \geq 0.81$, $RMSE_{cv} \leq 0.11$, $RER_{cv} \geq 9.85$, $RPD_{cv} \geq 2.31$). However, predictive models for total solids, lactose and ionic calcium showed limited accuracy ($R^2_{cv} \leq 0.59$, $RMSE_{cv} \geq 0.10$, $RER_{cv} \leq 7.17$, $RPD_{cv} \leq 1.54$), indicating the need for further investigation. **Industrial Implications:** Overall, this study bridges the gap between lab-scale NIR method evaluation and industrial implementation by validating the approach using samples from large-scale silos, marking an important step towards rapid, real-time compositional analysis in dairy processing. © 2026 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Funding Source: DPTC 2

Research Programme: ECF

A qualitative risk ranking approach of chemical contaminants for industrial needs: A case study on milk and dairy products

Wang X.; Monahan C.; Cummins E.

International Dairy Journal, 2025

Milk and dairy products are an important part of a healthy diet for many. Like all food products, they require constant vigilance for existing and emerging contaminants. Monitoring chemical contamination and managing the associated risks have become a top priority for dairy companies. Traditional risk ranking methods are mainly research-focused, requiring the understanding of complex numerical terms and extensive calculations. Therefore, in this study, a qualitative risk ranking approach was developed for companies to conduct a simple and quick prioritisation of the most significant hazards requiring investigation or interventions. To ensure product safety for health benefits and regulatory compliance, both health risk and policy risk were assessed following decision trees. Subsequently, chemicals were categorised into Tier 1 (RED), Tier 2 (AMBER), or Tier 3 (GREEN). Case studies demonstrate the practical applicability of the method through the analysis of literature data on three types of chemical hazards: established hazards (Aflatoxin M1), risk re-evaluation (Bisphenol A), and emerging contaminants (Per- and Polyfluorinated substances). The method provides a preliminary screening demanding a careful interpretation of results by users. Overall, the qualitative approach assists individual companies to use their own data and monitoring information to classify and prioritise chemical hazards. © 2025 The Authors

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DPTC 2 Publications: RFA 2

Funding Source: DPTC 2

Research Programme: RFA2

Highly selective trace ammonium removal from dairy wastewater streams by aluminosilicate materials

O'Connor E.; Kavanagh O.N.; Chovan D.; Madden D.G.; Cronin P.; Albadarin A.B.; Walker G.M.; Ryan A.

Journal of Industrial and Engineering Chemistry, 2020

Water is a key solvent, fundamental to supporting life on earth. It is equally important in many industrial processes, particularly within agricultural and pharmaceutical industries, which are major drivers of the global economy. The results of water contamination by common activity in these industries is well known and EU Water Quality Directives and Associated Regulations mandate that NH_4^+ concentrations in effluent streams should not exceed 0.3 mg L^{-1} , this has put immense pressure on organisations and individuals operating in these industries. As the environmental and financial costs associated with water purification begin to mount, there is a great need for novel processes and materials (particularly renewable) to transform the industry. Current solutions have evolved from combating toxic sludge to the use of membrane technology, but it is well known that the production of these membrane technologies creates a large environmental footprint. Zeolites could provide an answer; their pore size and chemistry enable efficient removal of aqueous based cations via simple ion exchange processes. Herein, we demonstrate efficient removal of NH_4^+ via both static and dynamic methodology for industrial application. Molecular modelling was used to determine the cation–framework interactions which will enable customisation and design of superior sorbents for NH_4^+ capture in wastewater. © 2019 The Korean Society of Industrial and Engineering Chemistry

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Funding Source: DPTC 2

Research Programme: RFA2

Microbial Community Redundancy and Resilience Underpins High-Rate Anaerobic Treatment of Dairy-Processing Wastewater at Ambient Temperatures

Paulo L.M.; Castilla-Archilla J.; Ramiro-Garcia J.; Escamez-Picón J.A.; Hughes D.; Mahony T.; Murray M.; Wilmes P.; O'Flaherty V.

Frontiers in Bioengineering and Biotechnology, 2020

High-rate anaerobic digestion (AD) is a reliable, efficient process to treat wastewaters and is often operated at temperatures exceeding 30°C, involving energy consumption of biogas in temperate regions, where wastewaters are often discharged at variable temperatures generally below 20°C. High-rate ambient temperature AD, without temperature control, is an economically attractive alternative that has been proven to be feasible at laboratory-scale. In this study, an ambient temperature pilot scale anaerobic reactor (2 m³) was employed to treat real dairy wastewater in situ at a milk processing plant, at organic loading rates of 1.3 ± 0.6 to 10.6 ± 3.7 kg COD/m³/day and hydraulic retention times (HRT) ranging from 36 to 6 h. Consistent high levels of COD removal efficiencies, ranging from 50 to 70% for total COD removal and 70 to 84% for soluble COD removal, were achieved during the trial. Within the reactor biomass, stable active archaeal populations were observed, consisting mainly of Methanotrix (previously Methanosaeta) species, which represented up to 47% of the relative abundant active species in the reactor. The decrease in HRT, combined with increases in the loading rate had a clear effect on shaping the structure and composition of the bacterial fraction of the microbial community, however, without affecting reactor performance. On the other hand, perturbances in influent pH had a strong impact, especially when pH went higher than 8.5, inducing shifts in the microbial community composition and, in some cases, affecting negatively the performance of the reactor in terms of COD removal and biogas methane content. For example, the main pH shock led to a drop in the methane content to 15%, COD removals decreased to 0%, while the archaeal population decreased to ~11% both at DNA and cDNA levels. Functional redundancy in the microbial community underpinned stable reactor performance and rapid reactor recovery after perturbations. © Copyright © 2020 Paulo, Castilla-Archilla, Ramiro-Garcia, Escamez-Picón, Hughes, Mahony, Murray, Wilmes and O'Flaherty.

Link to article: <https://doi.org/10.3389/fbioe.2020.00192>

Funding Source: DPTC 2

Research Programme: RFA2

Enhanced Methanization of Long-Chain Fatty Acid Wastewater at 20°C in the Novel Dynamic Sludge Chamber–Fixed Film Bioreactor

Singh S.; Holohan B.C.; Mills S.; Castilla-Archilla J.; Kokko M.; Rintala J.; Lens P.N.L.; Collins G.; O’Flaherty V.

Frontiers in Energy Research, 2020

Lipid-containing wastewaters, such as those arising from dairy processing, are frequently discharged at temperatures $\leq 20^{\circ}\text{C}$. Their valorization at low ambient temperatures offers opportunities to expand the application of high-rate anaerobic wastewater treatment toward achieving energy neutrality by minimizing the energy demand for heating. Lipid hydrolysis generates long-chain fatty acids (LCFAs), which incur operational challenges and hinder stable bioreactor operation by inducing sludge flotation and washout, coupled with the added challenge of treatment at lower temperature (20°C). These challenges are tackled together uniquely during the treatment of LCFA-rich synthetic dairy wastewater (SDW) (33% COD-LCFA) through de novo formed microbial granular sludge within the dynamic sludge chamber–fixed film (DSC-FF) reactor. The novel reactor design facilitated sludge retention for the entire operational period of 150 days by containing settled, flotating, and LCFA-encapsulated granular sludge and biofilm within a single module. High COD removal efficiencies (87–98%) were achieved in the three replicated DSC-FF reactors, along with complete LCFA removal at 18–72 h HRT (LCFA loading rate of 220–890 mgCOD-LCFA/L·day) and partial LCFA removal at 12 h HRT (LCFA loading rate of 1333 mgCOD-LCFA/L·day). The high removal efficiencies of unsaturated and saturated LCFAs achieved are reported for the first time during continuous anaerobic wastewater treatment at low temperatures (20°C). Moreover, de novo granulation was achieved within 8 days from a combination of inoculum mixtures at a high LCFA concentration (33% COD-LCFA) in SDW. The results demonstrate the feasibility of the DSC-FF reactor for treating LCFA-rich wastewaters at discharge temperatures and offer potential for expanded and more energetically productive anaerobic valorization of lipid-rich wastewater. © Copyright © 2020 Singh, Holohan, Mills, Castilla-Archilla, Kokko, Rintala, Lens, Collins and O’Flaherty.

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Funding Source: DPTC 2

Research Programme: RFA2

Milk reception in a time-efficient manner: A case from the dairy processing plant

Zacharski K.A.; Burke N.; Adley C.C.; Hogan P.; Ryan A.; Southern M.

Food Control, 2021

The short raw milk lifespan is a matter of concern for the dairy processing sector. It is crucial to the final product quality to ensure that raw milk will reach the cooling facility without undue delay while maintaining high hygienic standards. However, an effective milk reception is undermined by numerous internal and external challenges due to the complexity of the dairy manufacturing system and the stochastic variation of the milk supply chain. This work presents an industrial case study where the milk reception performance was examined, and opportunities for improvement were identified. The output provided operational documentation of each stage of milk reception. The outcomes illustrate that existing infrastructure and operations were not set up to manage a post-quota abolition uptake in milk production. A number of shortcomings and challenges were outlined, namely process bottlenecks, inefficient design of the facility layout, lack of standardized procedures, and internal-communication issues. Recommendations for improvements have been provided to achieve a 23.4% reduction of process lead time. These findings provide an opportunity for the industry to review their milk reception operations to deal with stochastic variations in milk supply and seasonality. © 2021 The Authors

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Funding Source: DPTC 2

Research Programme: RFA2

Fabrication and Evaluation of Filtration Membranes from Industrial Polymer Waste

Bano S.; Pednekar M.; Rameshkumar S.; Borah D.; Morris M.A.; Padamati R.B.; Cronly N.

Membranes, 2023

Polyvinylidene fluoride (PVDF) polymers are known for their diverse range of industrial applications and are considered important raw materials for membrane manufacturing. In view of circularity and resource efficiency, the present work mainly deals with the reusability of waste polymer 'gels' produced during the manufacturing of PVDF membranes. Herein, solidified PVDF gels were first prepared from polymer solutions as model waste gels, which were then subsequently used to prepare membranes via the phase inversion process. The structural analysis of fabricated membranes confirmed the retention of molecular integrity even after reprocessing, whereas the morphological analysis showed a symmetric bi-continuous porous structure. The filtration performance of membranes fabricated from waste gels was studied in a crossflow assembly. The results demonstrate the feasibility of gel-derived membranes as potential microfiltration membranes exhibiting a pure water flux of 478 LMH with a mean pore size of ~0.2 μm . To further evaluate industrial applicability, the performance of the membranes was tested in the clarification of industrial wastewater, and the membranes showed good recyclability with about 52% flux recovery. The performance of gel-derived membranes thus demonstrates the recycling of waste polymer gels for improving the sustainability of membrane fabrication processes. © 2023 by the authors.

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Funding Source: DPTC 2

Research Programme: RFA2

Optimising the quality management system in dairy processing

Burke, N.

Book: Quality control and Quality Assurance- Techniques and Applications, 2023

Milk has a solid reputation as a staple food since time immemorial. It is a complete food in its raw form, high in fat, protein, vitamins and minerals, including calcium. While the most beneficial first food for mammals is mammalian milk until weaning, cow's milk and dairy derivatives are considered significant nutritional components in the human diet. While milk consumption has in fact sharply declined in recent decades, the consumption of liquid milk derivatives and dairy products has steadily increased. Quality in terms of product, process and the environment in a milk production plant can be measured through performance, reliability and durability. The quality management system, in whatever form that may take within a plant, is the pinnacle in ensuring how one organisation can differentiate from its competitors. Quality systems and analytical testing protocols, especially in the dairy industry, are seldom quantified or fine-tuned to guarantee their efficiency. Furthermore, the impacts of quality systems on process, product, and environmental optimisation are frequently overlooked. This chapter reviews the activities that allow for the optimisation of quality systems in a dairy processing environment. The outcomes of which highlight the importance of process based quality systems.

Link to article: <https://doi.org/10.5772/intechopen.114055>

Funding Source: DPTC 2

Research Programme: RFA2

Enhancement of biomethane potential of brown sludge by pre-treatment using vortex based hydrodynamic cavitation

Islam M.S.; Ranade V.V.

Heliyon, 2023

Novel, non-thermal and economically benign pre-treatment process was developed for enhancing valorisation potential of brown sludge generated by dairy industry wastewater treatment plant (WWTP). Vortex-based hydrodynamic cavitation (HC) device was used to quantify influence of pretreatment by measuring biomethane potential (BMP) of untreated and treated brown sludge. Pre-treatment parameters, primarily, pressure drop and number of passes through the cavitation device were varied to quantify influence on BMP. BMP tests were performed at 39 °C containing 5% of total solids in each reactors using an automatic BMP measurement system containing 15 reactors with each volume of 500 mL fitted with overhead stirrer. HC treatment increased the soluble chemical oxygen demand (sCOD) by more than 25% which increased the BMP. HC treatment was able to push the BMP of treated sludge to more than 80% of the theoretical BMP. Volatile solids (VS) removal was more than 65%. Highest methane yield was 376 mL/g-VS of sludge. The methodology and results presented here show significant potential to valorise brown dairy sludge via vortex based hydrodynamic cavitation. © 2023 The Authors

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Funding Source: DPTC 2

Research Programme: RFA2

Hydrothermal carbonization of milk/dairy processing sludge: Fate of plant nutrients

Kwapinska M.; Pisano I.; Leahy J.J.

Journal of Environmental Management, 2023

Dairy processing sludge (DPS) is a byproduct generated in wastewater treatment plants located in dairy (milk) processing companies (waste activated sludge). DPS presents challenges in terms of its management (as biosolids) due to its high moisture content, prolonged storage required, uncontrolled nutrient loss and accumulation of certain substances in soil in the proximity of dairy companies. This study investigates the potential of hydrothermal carbonization (HTC) for recovery of nutrients in the form of solid hydrochar (biochar) produced from DPS originating from four different dairy processing companies. The HTC tests were carried out at 160 °C, 180 °C, 200 °C and 220 °C, and a residence time of 1h. The elemental properties of hydrochars (biochars), the content of primary and secondary nutrients, as well as contaminants were examined. The transformation of phosphorus in DPS during HTC was investigated. The fraction of plant available phosphorus was determined. The properties of hydrochar (biochar) were compared against the European Union Fertilizing Products Regulation. The findings of this study demonstrate that the content of nutrient in hydrochars (biochars) meet the requirements for organo-mineral fertilizer with nitrogen and phosphorus as the declared nutrients (13.9–26.7%). Further research on plant growth and field tests are needed to fully assess the agronomic potential of HTC hydrochar (biochar). © 2023 The Authors

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Funding Source: DPTC 2

Research Programme: RFA2

Release of N containing compounds during pyrolysis of milk / dairy sludge - preliminary experiential results

Kwapinska M.; Sommersacher P.; Kienzl N.; Retschitzegger S.; Lagler J.; Horvat A.; Leahy J.J.

Journal of Analytical and Applied Pyrolysis, 2023

A dried dairy processing sludge (sludge from wastewater treatment of an effluent from a milk processing plant) was pyrolysed in a single-particle reactor at different temperatures from 400 °C to 900 °C. NH₃ and HCN were measured online and offline by means of FTIR as well as by cumulative sampling in impinger bottles (in 0.05 M H₂SO₄ and 1 M NaOH, respectively) and analysed by photometric method. NO and NO₂ were measured online using a nitric oxide analyser while N₂O was measured by FTIR. Nitrogen (N) in the sludge and in the remaining char, char-N, was determined. Moreover, tar content in pyrolysis gas was measured and tar-N was determined. The results with respect to N mass balance closure are discussed. The different measurements techniques are compared. For pyrolysis at 520 °C and 700 °C nitrogen in the gas phase was mainly contained as N₂ (36 % and 40 % respectively), followed by NH₃ (15 % and 18 %), tar-N (10 % and 9 %), HCN (1 % and 3 %), NO (1 %) and NO₂ (0.2 %). The dairy processing sludge has very specific properties with organic-N present predominantly as proteins and a high content of inherent Ca. These characteristics affected the distribution of N. The amount of char-N was higher while the amount of tar-N lower than for sewage sludge from literature, at comparable pyrolysis temperature.

Link to article: <https://doi.org/10.1016/j.jaap.2024.106391>

Funding Source: DPTC 2

Research Programme: RFA2

Effects of food to inoculum ratio and ultrasound pre-treatment on biogas production from dissolved air flotation waste from dairy wastewater.

Liu, Y.; O'Connor, S.; Paulo, L.; Braguglia, C.; Gagliano, M.; O'Flaherty, V.

Process Safety & Environmental Protection, 2023

Dissolved air flotation (DAF) waste, a byproduct with high lipid content separated from dairy wastewater, is disposed of by land spreading and causing environmental pollution. To develop a sustainable treatment for DAF waste, this study investigated the effects of food to inoculum (F/I) ratio and ultrasound pre-treatment on the anaerobic digestion of DAF waste. The biochemical methane potential (BMP) of tested DAF waste ranged from 436–566 mL CH₄/g VS_{fed}. Increasing the F/I ratio (>1.0) inhibited methane production due to long chain fatty acids (LCFA) accumulation, which high concentrations of oleate inhibited methanogenesis and delayed palmitate degradation. Ultrasound pre-treatment with 30 min pulse (10 s on/ 10 s off) and 15 min continuous operations increased soluble chemical oxygen demand in DAF waste by 82% and 52%, respectively. Moreover, continuous sonication removed 38% of LCFA due to the implosion of cavitation bubbles. The BMP of DAF waste increased by 36% after sonication with F/I ratio 3.0. However, the lag time of methane production was prolonged after sonication due to the fast release of LCFA to the bulk solution. By implementing ultrasound pre-treatment and optimizing the F/I ratio, the energy potential of DAF waste can be harnessed, leading to more sustainable practices in dairy production.

Link to article: <https://doi.org/10.1016/j.psep.2024.01.058>

Funding Source: DPTC 2

Research Programme: RFA2

Psychrophilic and mesophilic anaerobic treatment of synthetic dairy wastewater with long chain fatty acids: Process performances and microbial community dynamics

Liu, YC; Ramiro-Garcia, J; Paulo, LM; Braguglia, CM; Gagliano, MC; O'Flaherty, V

BIORESOURCE TECHNOLOGY, 2023

Facilitating the anaerobic degradation of long chain fatty acids (LCFA) is the key to unlock the energy potential of lipids-rich wastewater. In this study, the feasibility of psychrophilic anaerobic treatment of LCFA-containing dairy wastewater was assessed and compared to mesophilic anaerobic treatment. The results showed that psychrophilic treatment at 15 °C was feasible for LCFA-containing dairy wastewater, with high removal rates of soluble COD (>90%) and LCFA (~100%). However, efficient long-term treatment required prior acclimation of the biomass to psychrophilic temperatures. The microbial community analysis revealed that putative syntrophic fatty acid bacteria and *Methanocorpusculum* played a crucial role in LCFA degradation during both mesophilic and psychrophilic treatments. Additionally, a fungal-bacterial biofilm was found to be important during the psychrophilic treatment. Overall, these findings demonstrate the potential of psychrophilic anaerobic treatment for industrial wastewaters and highlight the importance of understanding the microbial communities involved in the process.

Link to article: <http://dx.doi.org/10.1016/j.biortech.2023.129124>

Funding Source: DPTC 2

Research Programme: RFA2

Enhancing BMP and digestibility of DAF sludge via hydrodynamic cavitation

Islam M.S.; Ranade V.V.

Chemical Engineering and Processing - Process Intensification, 2024

This study addresses fats, oils, and grease bioconversion challenges in dissolved air floatation (DAF) sludge from dairy processing waste streams. A hydrodynamic cavitation (HC) based pre-treatment method was developed for enhanced anaerobic digestion of DAF sludge. Bench-scale pre-treatment experiments were carried out using a vortex-based HC device at 20 L/min and 250 kPa pressure drop. Influence of severity of the pre-treatment (number of passes through HC device) and substrate concentration on biomethane potential (BMP) was quantified. The study revealed that soluble chemical oxygen demand (sCOD) increases with number of passes (34 % after 80 passes). Higher sludge concentration was found to increase the lag time observed in the BMP data. It was possible to tolerate 3 % VS sludge. The pre-treatment showed a notable increase in BMP, exceeding 82 % of theoretical BMP, with VS removal surpassing 73 %. The highest methane yield achieved was 756 mL/gVS of sludge. The net energy gain (after subtracting energy required for pre-treatment) was found to be more than 100 kWh/ton of sludge. The developed pre-treatment process and presented results provide a basis for the effective valorisation of DAF sludge, promoting a circular economy approach. © 2024 The Author(s)

Link to article: <https://doi.org/10.1016/j.cep.2024.109733>

Funding Source: DPTC 2

Research Programme: RFA2

Enhanced biomethane production via hydrodynamic cavitation pretreatment and co-digestion of brown and DAF sludge

Islam M.S.; Ranade V.V.

Water Science and Technology, 2025

This study introduces a co-digestion and vortex-based hydrodynamic cavitation (HC) pretreatment of brown and dissolved air flotation (DAF) sludge derived from dairy processing waste streams to enhance biomethane potential (BMP). Co-digestion experiments were carried out for varying ratios of brown-to-DAF sludge (1:1, 2%; 3:1, 4%). The BMP tests, conducted at 39 °C using a 15-reactor system with a 400 mL reaction volume and overhead stirrers, demonstrated that HC treatment elevated soluble chemical oxygen demand (sCOD) of brown and DAF sludges by over 34% after 80 passes through HC device. Combining DAF with brown sludge improved methane yields and reduced the time to reach peak BMP. Loading 2% volatile solids (VS) with a 1:1 ratio resulted in methane production exceeding 90% of theoretical BMP, surpassing individual brown and DAF sludge BMPs (73 and 84%, respectively). Maximum VS removal surpassed 85%, achieving a peak methane yield of over 717 mL/g-VS. These findings offer insights into addressing effluent treatment challenges and highlight the potential of co-digestion for enhanced waste valorisation. © 2025 The Authors.

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Funding Source: DPTC 2

Research Programme: RFA2

Hydrothermal carbonization and pyrolysis of dairy processing sludge for improved nutrient management in agriculture: Current state-of-the-art

Kwapińska M.; Burke N.; Leahy J.J.

Chemical Engineering Journal Advances, 2025

This paper summarises advances in hydrothermal carbonization and pyrolysis of dairy processing sludge. Since municipal wastewater treatment plants are the primary source of sludge, thermochemical treatment processes were initially studied and applied to municipal sewage sludge. Research on dairy processing sludge can build upon the insights gained from sewage sludge studies. This article begins with a summary of the current state of the art for both technologies as applied to sewage sludge. It then presents an overview of dairy processing sludge properties, which in some cases differ significantly from those of sewage sludge. This review discusses the properties of all products derived from hydrothermal carbonization and pyrolysis of dairy processing sludge, identifies knowledge gaps, and explores the potential for integrating these two processes within wastewater treatment plants in dairy processing facilities. The integration of hydrothermal carbonization and pyrolysis presents a transformative approach to valorizing dairy processing sludge, aligning with global goals for climate resilience, nutrient recycling, and sustainable agriculture. The review highlights critical knowledge gaps and proposes strategies to accelerate industrial adoption and policy integration. © 2025 The Author(s)

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Funding Source: DPTC 2

Research Programme: RFA2

Functionalized organosolv lignin grafted with 3-aminopropyltriethoxysilane: A bio-based adsorbent for phosphate recovery from dairy wastewater

Masliha M.; Padnekar M.; De Micco J.; Ponnupandian S.; Mondal K.; Padamati R.B.

Heliyon, 2025

Wastewater rich in phosphates and nitrates causes eutrophication and leads to the impairment of freshwater resources. Out of various methods used, adsorption is the immaculate and economical for removing and recovering phosphates and nitrates from wastewater streams in a single-step process. This study explores the potential of bio-based adsorbent, functionalized organosolv lignin [OL], chemically modified by grafting with 3-aminopropyltriethoxysilane [APTES], as an effective bio-based adsorbent [OL-APTES-H⁺] for phosphate recovery from aqueous solutions and industrial wastewater. The characterization of OL-APTES-H⁺ was performed using multiple analytical techniques, providing comprehensive information on the material morphology, elemental composition, functional groups, thermal stability, surface charge, and electrokinetic behavior. The adsorption efficiency of OL-APTES-H⁺ was assessed under varying experimental conditions, including pH, contact time, and initial phosphate concentration. The adsorption capacity of OL-APTES-H⁺ depended on pH, with different forms of phosphate species being preferentially adsorbed at different pH values. A maximum adsorption capacity of 21.12 mg/g was achieved at pH 5. Kinetic studies indicated that the adsorption process followed a combination of electrostatic interactions, chemisorption and surface interaction, as evidenced by SEM and EDS analyses. XPS results confirm phosphorus incorporation on the adsorbent surface, reinforcing chemisorption. Adsorption isotherm analysis revealed that the data fitted well to the Langmuir isotherm model, suggesting a monolayer adsorption mechanism. The adsorption performance of OL-APTES-H⁺ was enhanced in the presence of monovalent ions, while a slight reduction in efficiency was observed in the presence of divalent anions. When applied to industrial dairy wastewater, OL-APTES-H⁺ exhibited phosphate removal efficiencies ranging from 30 % to 58 %. Overall, OL-APTES-H⁺ demonstrates considerable potential as a bio-based adsorbent for phosphate recovery, effectively mitigating environmental pollution in wastewater bodies and providing an eco-friendly source of phosphates for sustainable agricultural practices. © 2025 The Authors

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Funding Source: DPTC 2

Research Programme: RFA2

Valorizing Primary Digestate via Codigestion with DAF Sludge and Hydrodynamic Cavitation

Nayak J.K.; Islam M.S.; Ranade V.V.

Industrial and Engineering Chemistry Research, 2026

Anaerobic digestion (AD) efficiently converts biodegradable feedstocks to biogas; however, residual digestate remains challenging to valorize. Digestate from primary digesters retains significant carbon; however, it has a lower-than-desirable carbon-to-nitrogen (C/N) ratio (~7) and therefore typically yields low methane productivity. In this note, we present two strategies to enhance the methane yield from digestate. The first is to adjust the (C/N) ratio by mixing dissolved air flotation (DAF) sludge from the dairy industry, which has a high (C/N) ratio (~30). The DAF sludge is rich in fats, oils, and grease and is difficult to digest (operational difficulties, long lag phase, and poor biogas yields). In this study, we demonstrate the synergistic effect of the codigestion of primary digestate and DAF sludge. In this study, BMP tests were conducted on mixtures of primary digestate and DAF sludge to evaluate synergistic effects, defined as the ratio between the experimentally observed BMP of the mixture and the volatile solid (VS) weighted sum of BMPs obtained from the monodigestion of the individual substrates. A 1:1 blend (on a VS basis) exhibited the highest performance, achieving a synergy factor of 1.7 and a BMP of 684 mL CH₄/g VS. We also demonstrate that for cases where measured BMP is much smaller than the theoretical BMP (BMP_{th}), the HC pretreatment enabled methane recovery up to ~80% of the BMP_{th}. In this work, we used a vortex-based HC device for the pretreatment of a 1:1 mixture of digestate and DAF sludge and showed that the pretreated mixture could achieve 82% BMP_{th}. This HC-assisted codigestion approach provides a sustainable pathway for valorizing two of the most difficult waste streams in the AD and dairy industries, supporting renewable energy recovery and circular bioeconomy goals. © 2026 The Authors. Published by American Chemical Society

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DPTC 1 Publications: Pillar 1

Funding Source: DPTC 1

Research Programme: Pillar 1

Investigating the current skim milk powder inspection strategies for improvements in process optimisation

Burke N.; Southern M.; Ryan A.; Adley C.C.

Food Control, 2018

The production of skim milk powder (SMP) has increased among Irish Dairy processors in recent years. SMP has nutritional benefits and functional properties, including high calcium and potassium, a low fat content, excellent gelation, emulsification and foaming properties. As Irish dairy exports are increasing year on year, the requirement to intensify process optimisation is heightened. This study investigated the analytical test levels applicable to the regulatory requirements for the quality and safety assurance in the production of SMP across a 12 month period in two large Irish milk processing plants. The efficacy of the testing strategies and turnaround time for SMP for both plants were examined and compared. The results of this study demonstrate excess testing at 64% and 36% respectively, over the regulatory requirements for both companies. The sediment and density tests demonstrated the highest out of specification excess test results for each plant respectively. This is the first study to our knowledge on optimising sampling regimes in a dairy processing setting and the outcomes of this study propose an overview to the current SMP testing strategies for improvement in process optimisation for any milk product, focusing on the process in order to optimise overall production levels and to address analytical test levels, out of specification results and laboratory turn-around time (TAT). © 2018

Link to article: <https://doi.org/10.1016/j.foodcont.2018.06.025>

Funding Source: DPTC 1

Research Programme: Pillar 1

Evaluation of an environmental monitoring program for the microbial safety of air and surfaces in a dairy plant environment

Zacharski K.A.; Southern M.; Ryan A.; Adley C.C.

Journal of Food Protection, 2018

Microbiological hazards can occur when foodstuffs come into contact with contaminated surfaces or infectious agents dispersed by air currents in the manufacturing environment. An environmental monitoring program (EMP) is a critical aspect of sustainable and safe food manufacturing used to evaluate the effectiveness of the microbial controls in place. An effective EMP should be based on risk analysis, taking into account previous sampling history to determine the selection of the sampling points, the scope of the test, and the frequency of analysis. This study involved evaluation of the environmental monitoring regime and microbiological status of a medium-sized dairy plant manufacturing food ingredients, e.g., proteins, milk powders, and dairy fats. The data specific to microbial tests (n = 3,468), recorded across 124 fixed sampling locations over a 2-year period (2014 to 2015) from air (n = 1,787) and surfaces (n = 1,681) were analyzed. The aim of this study was to highlight the strengths and weaknesses of the EMP in a select dairy processing plant. The results of this study outline the selection of sampling locations, the scope of the test, and the frequency of analysis. An analysis of variance revealed subsections of the manufacturing areas with high risk factors, especially the packaging subsection specified for bulk packaging, the atomizer, and the fluidized bed. The temporal and spatial analysis showed the potential to reduce or relocate the monitoring effort, most notably related to total coliforms and *Staphylococcus aureus*, across the dairy plant due to homogeneity across the sampling subsections with little or no deviations. The results suggest a need to reevaluate the current EMP and the corrective action plan, especially with regard to detection of pathogens. Recommendations for optimization of the EMP are presented to assist the dairy industry with reviewing and revising the control measures and hazard assessment with regard to existing contamination issues. Copyright © 2018 International Association for Food Protection.

Link to article: <https://doi.org/10.4315/0362-028X.JFP-17-464>

Funding Source: DPTC 1

Research Programme: Pillar 1

Modelling & Simulation as a Strategic Tool for Decision-Making Process: A Dairy Case Study

Eccher C.; Geraghty J.

Decision Making in Manufacturing and Services, 2020

The Dairy Industry faces many challenges compared to other sectors. On the supply side due to the nature of the raw material, large inventories are not applied; during the manufacturing process, the continuous production is highly sensitive to any sort of unplanned disruption; and on the demand side, the market dictates the commodity prices. In response to the growth in competition, dairy organizations' strategy must incorporate technology into their daily processes in order to become more efficient, profitable and sustainable. To achieve desired levels of improvement, Modelling and Simulation has been increasing in popularity in the decision-making process. Using a Dairy company as a case study, this paper has highlighted the potential for Modelling and Simulation to be used as a powerful strategic tool for decision-making processes.

Link to article: <https://doi.org/10.7494/dmms.2020.14.1.2785>

Funding Source: DPTC 1

Research Programme: Pillar 1

Evaluating the Temperature of Incoming Raw Milk at a Dairy Processing Plant

Burke, N; Zacharski, K; Adley, C.C.; Southern, M.

HSOA Journal of Dairy Research and Technology, 2021

Ensuring stringent temperature control parameters and efficient cooling mechanisms are maintained at a dairy plant are critical to downstream processing capabilities and product quality. This study examined the temperature of incoming raw milk across three production months (June, July, August 2018), to capture peak season processing at one dairy manufacturing plant. A total of n=4,236 data points across three milk intake bays were analysed. Across the study area, the average temperature of incoming milk was 6.18°C (1.54 Standard Deviation (SD)). After the initial cooling step in the dairy facility, the average temperature of stored raw milk was 4.74°C (0.77 SD), with an average cooling performance of -1.44°C (SD 1.34). This research highlights for the first time, the analysis of a narrow but important quality step in a milk processing plant, namely the critical criteria of temperature monitoring and flow rate management at a dairy processing facility.

Link to article: <https://doi.org/10.24966/DRT-9315/100024>

DPTC 1 Publications: Pillar 2

Funding Source: DPTC 1

Research Programme: Pillar 2

Monitoring of pilot-scale induction processes for dairy powders using inline and offline approaches

O'Sullivan J.J.; Schmidmeier C.; Drapala K.P.; O'Mahony J.A.; Kelly A.L.

Journal of Food Engineering, 2017

The induction of two dairy powders, skim milk powder (SMP; low-protein content), and milk protein isolate (MPI, high-protein content), was studied. The powder induction approaches investigated were (1) eductor alone, (2) eductor with a static mixer, and (3) eductor with high shear inline mixing. Measurement of pressure drop, from which viscosity was determined inline using the Hagen-Poiseuille equation, offline viscometry and particle size analyses were performed. High shear inline mixing provided the most efficient induction of powders. In addition, more rapid powder induction, as observed from particle size analysis, was achieved for SMP in comparison to MPI, owing to its better rehydration properties. Inline pressure drop data demonstrated that dissolution of MPI had two distinct phases: (i) powder introduction, and (ii) powder breakdown, irrespective of configuration and concentration employed. © 2016 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.jfoodeng.2016.10.023>

Funding Source: DPTC 1

Research Programme: Pillar 2

Recent advances on microbial transglutaminase and dairy application

Romeih E.; Walker G.

Trends in Food Science and Technology, 2017

Background Microbial transglutaminase (MTGase) is an enzyme widely known to modify food proteins. MTGase is capable of forming both inter- and intra-molecular isopeptide bonds in various food proteins by cross-linking the amino acid residues of protein bound glutamine (acyle donor) and lysine (acyle receptor). The use of MTGase has increased for various dairy-based systems to promote and develop desired functional changes in dairy products. **Scope and approach** The specificity of MTGase towards milk proteins and the bioavailability of cross-linked proteins are key factors for industry and consumer preferences. Various attempts have been made to utilize MTGase to improve yield and to advance the functional properties of cheese types, fermented milks, milk powders, caseinate and other dairy products, which are discussed hereby. **Key findings and conclusions** The implication of MTGase in dairy protein-based products alters the technological/functional properties of milk proteins, including gelation, heat stability, viscosity, emulsification, water holding capacity and foaming stability. Variations in the reaction conditions such as enzyme concentration, temperature, pH, substrate availability and specificity enable varying grades of protein modification. MTGase is considered since 1998 as GRAS; generally recognized as safe, and is recognized as a safe substance for human intake. The frontiers of knowledge and technology in this review contribute substantially to establish a foundation for the dairy industry to develop innovative and/or functional product with satisfactory flavour and improved textural properties. © 2017 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 2

The case for milk protein standardisation using membrane filtration for improving cheese consistency and quality

Soodam K.; Guinee T.P.

International Journal of Dairy Technology, 2018

Milk composition varies with season owing to stage of lactation and variation in diet and weather. Variation in the concentration of milk protein is conducive to inconsistency in cheese yield, composition and quality especially where standard operating procedures are not objectively standardised with reference to casein content. Milk protein standardisation (e.g. to 4.5%) by low-concentration factor ultrafiltration (LCFUF) or microfiltration (LCFMF) provides an effective means of obtaining more consistent cheese manufacture. Consequently, LCFUF is now widely practised. The benefits of LCFUF and LCFMF and the effect of key processing parameters on the quality of cheese from protein-standardised milk are reviewed. © 2018 Society of Dairy Technology

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Funding Source: DPTC 1

Research Programme: Pillar 2

Effects of vibration parameters and pipe insertion depth on the motion of particles induced by vertical vibration

Zhang F.; Cronin K.; Lin Y.; Liu C.; Wang L.

Powder Technology, 2018

Granular particles can be induced to move against gravity upwards through a pipe or tube that is partially inserted under the powder free surface while subject to vertical vibration. This offers a new approach for transporting bulk material. In this paper, the effects of both vibration parameters and insertion depth of a pipe on particle motion are experimentally studied. A minimum vibration amplitude (A) and frequency (f) are necessary for particle motion to occur. There is a monotonic increase of the final rise height of the powder (h_{eq}) with increasing amplitude A . However, h_{eq} exhibits a non-monotonic dependence on frequency, f . There is an optimum frequency at which particles climb highest, and any further increase of frequency leads to a diminishment of this upwards motion. A phase diagram of particle movement is presented which shows that different zones of motion exist. This unique finding suggests that the mechanism of particle movement is caused by the creation and filling of voids under the tube. Particles cannot move upwards when the pipe insertion depth, h_{in} is <1.5 mm irrespective of how strong the supplied vibration is. In general, increasing h_{in} can improve climbing, but this effect falls with increasing levels of insertion depth until a saturation level is reached. PACS number: 45.70.MG - granular flow © 2017 Elsevier B.V.

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Funding Source: DPTC 1

Research Programme: Pillar 2

The biological activity of fermented milk produced by *Lactobacillus casei* ATCC 393 during cold storage

Abdel-Hamid M.; Romeih E.; Gamba R.R.; Nagai E.; Suzuki T.; Koyanagi T.; Enomoto T.

International Dairy Journal, 2019

Lactobacillus casei ATCC 393 is an important probiotic strain widely known in dairy technology. However, its capability to produce bioactive peptides from milk proteins has not been studied. The viability of the *Lb. casei* ATCC 393 strain and some physicochemical properties in fermented milk throughout storage for 21 days at 4 °C was evaluated; biological activity, i.e., antioxidant, angiotensin converting enzyme inhibitory and anticancer activities of water soluble extract and its filtrate (< 2 kDa; F1) were determined. *Lb. casei* counts remained over $9 \log \text{cfu g}^{-1}$ during the storage period in fermented milk. These bioactivities were increased significantly ($P < 0.01$) during storage. F1 of fermented milk after three weeks of storage showed the highest bioactivity impact. De novo sequencing assay for peptide identification was applied to the mass spectrum of F1. The promising capability of *Lb. casei* ATCC 393 to release bioactive peptides from milk proteins was demonstrated. © 2018 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.idairyj.2018.12.007>

Funding Source: DPTC 1

Research Programme: Pillar 2

Influence of calcium-binding salts on heat stability and fouling of whey protein isolate dispersions

Hebishy E.; Joubran Y.; Murphy E.; O'Mahony J.A.

International Dairy Journal, 2019

The effect of the calcium-binding salts (CBS), trisodium citrate (TSC), tripotassium citrate (TPC) and disodium hydrogen phosphate (DSHP) at concentrations of 1–45 mM on the heat stability and fouling of whey protein isolate (WPI) dispersions (3%, w/v, protein) was investigated. The WPI dispersions were assessed for heat stability in an oil bath at 95 °C for 30 min, viscosity changes during simulated high-temperature short-time (HTST) and fouling behaviour using a lab-scale fouling rig. Adding CBS at levels of 5–30 mM for TSC and TPC and 25–35 mM for DSHP improved thermal stability of WPI dispersions by decreasing the ionic calcium (Ca^{2+}) concentration; however, lower or higher concentrations destabilised the systems on heating. Adding CBS improved heat transfer during thermal processing, and resulted in lower viscosity and fouling. This study demonstrates that adding CBS is an effective means of increasing WPI protein stability during HTST thermal processing. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.idairyj.2018.12.003>

Funding Source: DPTC 1

Research Programme: Pillar 2

Analysing extraction uniformity from porous coffee beds using mathematical modelling and computational fluid dynamics approaches

Moroney K.M.; O'Connell K.; Meikle-Janney P.; O'Brien S.B.G.; Walker G.M.; Lee W.T.

PLoS ONE, 2019

Achieving a uniform extraction of soluble material from a porous matrix is a generic problem in various separation and filtration operations, with applications in the food processing, chemical and pharmaceutical industries. This paper describes models of fluid flow and transport of soluble material within a packed granular bed in the context of coffee extraction. Coffee extraction is described by diffusion of soluble material from particles of one or more representative sizes into fluid flowing through the packed bed. One-dimensional flow models are compared to computational fluid dynamics (CFD) models. A fine and a coarse coffee grind are considered. Model results are compared to experimental data for a packed cylindrical coffee bed and the influence of a change in geometry to a truncated cone is considered. Non-uniform flow in the truncated cone causes significant variation in the local extraction level. Coffee extraction levels during brewing are analysed using extraction maps and the degree of variation is represented on the industry standard coffee brewing control chart. A high variation in extraction yield can be expected to impart bitter flavours into the brew and thus is an important variable to quantify. © 2019 Moroney et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Link to article: <https://doi.org/10.1371/journal.pone.0219906>

Funding Source: DPTC 1

Research Programme: Pillar 2

Influence of particle size on the physicochemical properties and stickiness of dairy powders

O'Donoghue L.T.; Haque M.K.; Kennedy D.; Laffir F.R.; Hogan S.A.; O'Mahony J.A.;
Murphy E.G.

International Dairy Journal, 2019

The compositional and physicochemical properties of different whey permeate (WPP), demineralised whey (DWP) and skim milk powder (SMP) size fractions were investigated. Bulk composition of WPP and DWP was significantly ($P < 0.05$) influenced by powder particle size; smaller particles had higher protein and lower lactose contents. Microscopic observations showed that WPP and DWP contained both larger lactose crystals and smaller amorphous particles. Bulk composition of SMP did not vary with particle size. Surface composition of the smallest SMP fraction ($<75 \mu\text{m}$) showed significantly lower protein (-9%) and higher fat ($+5\%$) coverage compared with non-fractionated powders. For all powders, smaller particles were more susceptible to sticking. Hygroscopicity of SMP was not affected by particle size; hygroscopicity of semi-crystalline powders was inversely related to particle size. This study provides insights into differences between size fractions of dairy powders, which can potentially impact the sticking/caking behaviour of fine particles during processing. © 2019 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 2

Pneumatic conveying of cohesive dairy powder: Experiments and CFD-DEM simulations

Olaleye A.K.; Shardt O.; Walker G.M.; Van den Akker H.E.A.

Powder Technology, 2019

We performed an experimental and numerical investigation of pneumatic conveying of cohesive dairy powder. The experiments with fat-filled milk powder (FFMP) fines with an average particle size of 94 μm were carried out in a 2-inch diameter stainless steel pipe consisting of two 2.5 m horizontal sections connected to a 0.65 m vertical section by two bends of 0.4 m radius each. In addition to measurements of pressure drop and powder deposition, an optical technique was used to measure the dynamics (probability densities) of local particle volume fractions as a function of operating conditions. Numerical simulations were performed with a commercial discrete element modelling (DEM) software, EDEM[®], coupled with the computational fluid dynamics (CFD) software, FLUENT[®]. The simulation results in terms of pressure drops and particle volume fractions were compared with the experimental data. A very satisfactory agreement was found. At low gas velocities, cohesive dairy powders easily re-agglomerate after the second 90° bend and then deposit at the bottom of the horizontal pipe. At higher gas velocities, results show intermittent dispersion of particles and less particle deposition is observed even at higher loading ratio. © 2019 Elsevier B.V.

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Funding Source: DPTC 1

Research Programme: Pillar 2

Atomisation technologies used in spray drying in the dairy industry: A review

O'Sullivan J.J.; Norwood E.-A.; O'Mahony J.A.; Kelly A.L.

Journal of Food Engineering, 2019

Atomisation is an integral element of the spray-drying process, whereby a bulk liquid feed is converted to discrete droplets, greatly increasing the surface area of the feed liquid and thereby increasing considerably the achievable rates of evaporation of water. These droplets, through evaporation of water in the main dryer chamber, become individual powder particles during the spray-drying process. This review provides a comprehensive examination of the most recent developments in atomisation technology for spray-drying, with a particular focus upon dairy applications (e.g., skim and whole milk powders, casein- and whey-based powders, and fat-filled milk powders). As well as a review of principles of different technologies for atomisation, such as rotary atomisers, pressure nozzle atomisers, pneumatic atomisers, ultrasonic atomisers and electrospray atomisers, the industrial applicability and challenges in use of each approach to atomisation are presented. Approaches for monitoring the atomisation process and other factors that influence atomisation, such as feed composition and key process parameters, are considered to provide a holistic analysis of the atomisation process. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 2

Sealing pipe top enhancing transportation of particulate solids inside a vertically vibrating pipe

Zhang F.; Cronin K.; Lin Y.; Miao S.; Liu C.; Wang L.

Powder Technology, 2019

Particles can move against gravity inside a vibrating tube inserted in a static granular bed. This offers a new approach for transporting bulk material. In this work, we demonstrate a method to enhance the conveying of powder by sealing the tube top. With the same vibration conditions, a comparison of particle motion in an opened tube and closed top (sealed) pipe is made. Compared to an un-sealed pipe, particle upward motion within a sealed pipe is improved. With low vibration strength, only particles in the sealed tube can ascend. With increasing vibration strength, particles can climb in both tubes while particles in sealed pipe move faster and higher. The enhancement effect works well for particles of smaller size ($d < 1$ mm), and the positive effect becomes weaker with an increase in particle diameter. In a sealed tube, the final height of the granular column increases as the tube length increases while the growth velocity is reduced. Particle conveying in sealed tube shows less dependence on tube diameter compared to an un-sealed tube. Sealing the tube top introduces air pressure difference during each vibration cycle, which induces an additional upward drag force on the particles in the tube. The drag force becomes significant compared to other relevant forces for small diameter particles at high levels of vibration. © 2018 Elsevier B.V.

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Funding Source: DPTC 1

Research Programme: Pillar 2

Influence of mechanical integrity during pneumatic conveying on the bulk handling and rehydration properties of agglomerated dairy powders

Hazlett R.; Schmidmeier C.; O'Mahony J.A.

Journal of Food Engineering, 2020

Agglomerated powders are susceptible to breakdown on handling, most notably, during powder conveying. In this study, three agglomerated dairy powders (whey protein concentrate powder, WPC; fat-filled milk powder, FFMP and infant formula powder, IF) were conveyed through a custom-fabricated dispersion rig to understand the effects of agglomerate breakdown on dairy powder handling and application. All samples displayed significant breakdown on dispersion, evidenced by reducing particle size and increasing bulk density. The resulting flowability of these powders was impaired (flow index: WPC: 9.3 to 5.1, FFMP: 5.7 to 4.9 and IF: 16 to 10) via increased particle-particle interactions. The initial stages of rehydration were impeded by agglomerate breakage (42.9–47.0% wettability reduction and 7.22–16.4% dispersibility reduction), while powder solubility remained relatively unchanged. This study provides insights into the alterations of agglomerated dairy powder properties on agglomerate breakdown, while identifying the effects these alterations have on the functional properties of these powders. © 2020 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 2

A preliminary evaluation of the impact of pulsed electric field and high-pressure processing treatments on mobility of norbixin molecules through rennet-induced casein matrices

Alehosseini A.; Wall C.; Segat A.; Tiwari B.K.; Sharma P.; Kelly A.L.; Sheehan J.J.

Journal of Food Process Engineering, 2021

Considering the turnover of global cheese industry and while colored cheese is a subset of this, the issue of producing colored cheese without compromising the quality and economic value of the whey stream is a significant one. In this work, the potential of two pilot-scale processes—pulsed electric field (PEF) and high-pressure processing (HPP)—to increase the diffusion rate of norbixin molecules through a casein model system, rennet-induced micellar casein concentrate, and Cheddar cheese, were evaluated independently. Varying PEF treatments (2.4 kV cm⁻¹, pulse widths: 5–7 μs) were applied to alter the degree of sample permeabilization; however, the migration patterns of norbixin molecules were not statistically affected. Cheddar cheese samples were also subjected to high pressure (HP) treatments (250–400 MPa, 10 min). Confocal laser scanning micrographs of the HP-treated samples showed more distinct spherical fat globules surrounded by the continuous protein phase, suggesting HP-treatment considerably assists hydration of the protein phase and induces the distinctive fat mass appearance. However samples L*, a*, and b* color values were not altered significantly other than minor colorant diffusion through surface crevices. While the operational parameters, at the range applied in this work, did not significantly increase the penetration rate of norbixin through the samples, the resulting findings do significantly add to the current knowledge and through further research should considerably shorten the path to achieving a practical solution. Practical Applications: The two industrially scalable approaches (i.e., PEF and HPP) that have been evaluated in the current research have not previously been applied to increase the migration of norbixin molecules through the renneted casein structure. Furthermore, the application of PEF to cheeses is one in which interest is now growing. The current authors believe that the current study has a high degree of commercial importance and could be used as a catalytic study for further research around manipulating the relevant parameters and optimizing the process, so that PEF and HPP can finally be used at industrial scale. © 2021 Wiley Periodicals LLC.

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Funding Source: DPTC 1

Research Programme: Pillar 2

The influence of temperature on filtration performance and fouling during cold microfiltration of skim milk

France T.C.; Bot F.; Kelly A.L.; Crowley S.V.; O'Mahony J.A.

Separation and Purification Technology, 2021

Changes in the physicochemical properties and distribution of constituents in skim milk during microfiltration (MF) at low temperature influence filtration performance and product composition. In this study, the influence of processing temperature within the cold MF range (4, 8 and 12 °C) on filtration performance, fouling and partitioning of proteins was investigated. MF at 4 °C required the greatest energy input due to the significantly higher ($p < 0.05$) viscosity of feed and retentate streams, compared to processing at 8 and 12 °C. The greatest and lowest extents of reversible and irreversible fouling during MF were observed on filtration at 12 and 4 °C, respectively. Chemical analysis of the cleaning solutions post-processing demonstrated that protein was the major foulant; the lowest protein content in the recovered cleaning solutions (50 °C water and 55 °C alkali) was measured after MF at 4 °C. The concentration of β -casein, β -lactoglobulin and α -lactalbumin in the permeate all decreased throughout MF, due to fouling of the membrane. The greatest decrease in concentration of β -casein in the permeate during MF was observed at 12 °C (18.1%) followed by 8 °C (17.1%) and 4 °C (13.6%). The results of this study provide valuable information on processing efficiency (i.e., energy consumption and protein yield) and membrane fouling during the processing of skim milk in the cold MF range. © 2021 Elsevier B.V.

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Funding Source: DPTC 1

Research Programme: Pillar 2

Approaches for improving the flowability of high-protein dairy powders post spray drying – A review

Hazlett R.; Schmidmeier C.; O'Mahony J.A.

Powder Technology, 2021

Challenges are commonly encountered in the bulk handling and application of high-protein dairy powders, and are strongly influenced by their poor flowability. Powder flowability can be defined as the ability of a powder to flow under set environmental or processing conditions and it is ultimately determined by the type and extent of interparticle interactions occurring in the bulk powder (e.g., van der Waals and electrostatic interactions). High-protein powders are particularly susceptible to the occurrence of interparticle interactions, resulting in increased cohesive forces being experienced in the bulk powder, thereby reducing powder flowability. This review summarises the major factors responsible for poor flowability in high-protein dairy powders and critiques traditional (e.g., agglomeration) and some of the more relevant novel approaches (e.g., dry- and wet-coating and roller compaction) available for improving the flowability of powders post-spray drying. This review material will be of considerable interest to dairy scientists, technologists and engineers challenged with understanding, predicting and controlling the bulk handling and flowability of high-value dairy protein powders. © 2021 The Authors

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Funding Source: DPTC 1

Research Programme: Pillar 2

Enzymatically cross-linked skim milk powder: Enhanced rheological and functional properties

Romeih E.; Albadarin A.B.; Olaleye A.; Walker G.

International Dairy Journal, 2021

Enzymatically cross-linking milk proteins by microbial transglutaminase (MTGase) is a promising strategy in developing and promoting desired multi-functional properties of dairy foods. This study evaluated the effect of MTGase treatments (0.5, 1, 2 and 4 U g⁻¹ milk protein) of skim milk at two different incubation conditions (20 min at 38 °C, 12 h at 4 °C) on rheological and structural characteristics of spray dried skim milk powders (SMP). SMP samples treated with MTGase showed significantly increased particle size. Cross-linking milk proteins by MTGase revealed significant improvements in powder flowability characteristics by means of lower aeration and basic flow energies, and higher bulk density and compressibility properties compared with the control SMP. Overall, cross-linked SMP offers a promising option as a multi-functional ingredient for the dairy and/or composite food producers, and appearing to be an efficient option in improving powder rheological properties for dairy powder manufacturers. © 2020 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 2

Two-stage valve homogenisation enhances particle dispersion in milk protein concentrates during reconstitution and reduces heat-induced particle aggregation in resultant dispersions

Hebishy E.; Le Berre M.; Crowley S.V.; O'Mahony J.A.

Frontiers in Food Science and Technology, 2022

Milk protein concentrates (MPCs) are highly functional ingredients, with high-protein variants increasingly used in numerous applications. The objective of this study was to determine the impact of homogenisation, as part of the rehydration process, on solubility and heat stability of MPC. An 80% protein MPC powder was reconstituted (3% protein, w/v) and homogenised at 50°C using a pilot-scale, two-stage, valve homogeniser at different total pressures of 0, 5, 10, 15 and 20 MPa. Rehydrated samples were analysed for solubility, particle size, protein profile and heat stability (change in particle size distribution on heating in an oil bath at 140°C for 5 min). The results showed a considerable increase in solubility after applying homogenisation at 5 MPa. Homogenisation at pressures of 5–10 MPa reduced particle size of MPC dispersions further, with further increases in pressure having no additional effect. Increased heat stability was observed on increasing homogenisation pressures up to 10 MPa. This work demonstrates the positive impact of homogenisation on particle dispersion in MPCs and identifies a possible link between improved dispersion and heat stability. This would be applied as a strategy in dairy plants to reduce fouling in heat surfaces which suggests a significant economic impact in dairy processing. Copyright © 2022 Hebishy, Le Berre, Crowley and O'Mahony.

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Funding Source: DPTC 1

Research Programme: Pillar 2

An Experimental Study on the Dilute Phase Pneumatic Conveying of Fat-Filled Milk Powders: Particle Breakage

Zhang F.; O'Mahony J.A.; Miao S.; Cronin K.

Powders, 2023

Powder breakage during pneumatic conveying negatively affects the properties of dairy products and causes increased dusting, reduced wettability, and decreased product performance. In particular, particle breakage is a serious issue for fat-filled milk powder (FFMP) which, if it breaks, releases fat that causes odours and leads to sticky blocked pipes. In this work, a conveying rig (dilute phase, positive pressure) with 50 mm diameter food grade stainless steel pipes (1.5 m high and 5 m conveying distance with three 90° bends, two in the vertical plane and one in the horizontal plane) was built as the test system. The effects of operating conditions (conveying air velocity and solid loading rate) on the attrition of FFMP in a dilute phase conveying system were experimentally studied. Four quality characteristics were measured before and after conveying: bulk density, particle size distribution, wettability, and solubility, to assess the influence of particle breakage. Conveying air speed shows a significant impact on powder breakage. As air speed increased, more breakage occurred, and the volume mean diameter $D[4,3]$ decreased by around 50%, using the largest conveying air speed of 38 m/s. Bulk density increased accordingly whereas wettability decreased with an increase in air speed, resulting from the higher breakage rate. On other hand, improving the solid loading rate can further reduce the breakage level, but the positive effect is not as good as decreasing air speed. © 2023 by the authors.

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DPTC 1 Publications: Pillar 3

Funding Source: DPTC 1

Research Programme: Pillar 3

Short communication: Multi-component interactions causing solidification during industrial-scale manufacture of pre-crystallized acid whey powders

Drapala K.P.; Murphy K.M.; Ho Q.T.; Crowley S.V.; Mulcahy S.; McCarthy N.A.; O'Mahony J.A.

Journal of Dairy Science, 2018

Acid whey (AW) is the liquid co-product arising from acid-induced precipitation of casein from skim milk. Further processing of AW is often challenging due to its high mineral content, which can promote aggregation of whey proteins, which contributes to high viscosity of the liquid concentrate during subsequent lactose crystallization and drying steps. This study focuses on mineral precipitation, protein aggregation, and lactose crystallization in liquid AW concentrates (~55% total solids), and on the microstructure of the final powders from 2 independent industrial-scale trials. These AW concentrates were observed to solidify either during processing or during storage (24 h) of pre-crystallized concentrate. The more rapid solidification in the former was associated with a greater extent of lactose crystallization and a higher ash-to-protein ratio in that concentrate. Confocal laser scanning microscopy analysis indicated the presence of a loose network of protein aggregates ($\leq 10 \mu\text{m}$) and lactose crystals (100–300 μm) distributed throughout the solidified AW concentrate. Mineral-based precipitate was also evident, using scanning electron microscopy, at the surface of AW powder particles, indicating the formation of insoluble calcium phosphate during processing. These results provide new information on the composition- and process-dependent physicochemical changes that are useful in designing and optimizing processes for AW. © 2018 American Dairy Science Association

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Funding Source: DPTC 1

Research Programme: Pillar 3

Impact of Residual Lactose on Dry Heat-Induced Pre-texturization of Whey Proteins

Gulzar M.; Jacquier J.C.

Food and Bioprocess Technology, 2018

Controlling denaturation/aggregation of whey proteins during their pre-texturization is highly critical to avoid variability in their functional properties. We investigated how the dry heat-induced (16 h at 100 °C and $a_w = 0.23$) pre-texturization of whey protein isolate (WPI) is affected by traces of remaining lactose (0.3–2.0%) and how it influences its subsequent gelling properties. Lactose even in trace quantities developed intense browning of WPI. Dry-heating conditions used in this study mainly developed soluble aggregates stabilized by covalent crosslinks other than disulfide bonds. The extent of aggregation and size of aggregates were drastically increased with increasing lactose. Intermediate quantity (39–46%) of soluble aggregates improved the gel strength, while excessive aggregation (> 50%) resulted in loss of gel strength. Elasticity of gels was also increased by increasing protein aggregates. This study suggests that the traces of lactose that remain in WPI are critical for controlling its pre-texturization by dry heating and its subsequent gelling properties. © 2018, Springer Science+Business Media, LLC, part of Springer Nature.

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Funding Source: DPTC 1

Research Programme: Pillar 3

Effect of pH and heat treatment on viscosity and heat coagulation properties of milk protein concentrate

Ho Q.T.; Murphy K.M.; Drapala K.P.; O'Callaghan T.F.; Fenelon M.A.; O'Mahony J.A.;
McCarthy N.A.

International Dairy Journal, 2018

The effect of pH, adjusted using either hydrochloric acid (HCl), citric acid or sodium hydroxide, on calcium ion (Ca^{2+}) activity, and consequent changes in viscosity and heat coagulation time (HCT) of milk protein concentrate (MPC) was investigated. Reducing the pH of MPC dispersions resulted in a reduction in their viscosity, which subsequently increased during heat treatment. The maximum heat stability of MPC was observed at pH 6.7. Reducing the pH of MPC from 6.7 to 6.2 resulted in a significant ($P < 0.05$) increase in Ca^{2+} activity, and reduction in HCT. Such changes were more extensive using HCl compared with citric acid. Increasing the pH greater than 6.7 also led to a reduction in HCT but a decrease in Ca^{2+} activity. These results demonstrate the importance of pH adjustment, and choice of acidulant, on Ca^{2+} activity, viscosity, and heat coagulation properties of MPC concentrates during processing. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Effect of 3D printing on the structure and textural properties of processed cheese

Le Tohic C.; O'Sullivan J.J.; Drapala K.P.; Chartrin V.; Chan T.; Morrison A.P.; Kerry J.P.; Kelly A.L.

Journal of Food Engineering, 2018

Three-dimensional (3D) printing is a process whereby complex three-dimensional objects are generated. In this study, 3D printing was investigated for food applications, using a commercially available processed cheese as the printing material. After melting at 75 °C for 12 min, the processed cheese was printed using a modified commercial 3D printer at low or high extrusion rates. Comparative assessment of untreated, melted and printed cheeses was conducted employing texture profile analysis, rheology, colourimetry and confocal laser scanning microscopy (CLSM). Processing (i.e., melting and extrusion) had a significant impact upon cheese properties. Melted and printed cheese samples were significantly ($P < 0.05$) less hard, by up to 49%, and both exhibited higher degrees of meltability, ranging from 14% to 21%, compared to untreated cheese samples. This shows that 3D printing substantially changes the properties of processed cheese, possibly offering new potential applications for tailoring structures using this novel process.

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Funding Source: DPTC 1

Research Programme: Pillar 3

Influence of protein standardisation media and heat treatment on viscosity and related physicochemical properties of skim milk concentrate

Murphy K.M.; Ho Q.T.; Drapala K.P.; Keena G.M.; Fenelon M.A.; O'Mahony J.A.; McCarthy N.A.

International Dairy Journal, 2018

The effects of heat treatment and protein standardisation on the physical properties of skim milk concentrates were determined. Protein standardisation was carried out by the addition of lactose or milk permeate to skim milk. Unstandardised and standardised skim milk was subjected to heat treatment temperatures of 90 or 120 °C prior to evaporation whereafter the solids content was increased to 46% (w/w). Viscosity data showed non-standardised concentrates had the highest viscosity, followed by skim standardised with milk permeate followed by that standardised with lactose. Thermal treatment at 120 °C also resulted in a higher viscosity than that at 90 °C for all concentrates. Particle size data of evaporated skim milk showed a bimodal size distribution for skim milk standardised with liquid milk permeate, compared with monomodal distribution profiles for unstandardised skim milk and lactose standardised skim milk. Overall, this study showed that protein standardisation and standardisation media significantly affected concentrate properties. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

The use of inline high-shear rotor-stator mixing for preparation of high-solids milk protein-stabilised oil-in-water emulsions with different protein:fat ratios

O'Sullivan J.J.; Drapala K.P.; Kelly A.L.; O'Mahony J.A.

Journal of Food Engineering, 2018

The emulsification of refined palm oil (RPO) in a continuous phase consisting of skim milk concentrate (SMC) and maltodextrin with a dextrose equivalent value of 17 (MD17) to produce fat-filled milk emulsions (FFMEs), was studied. A novel inline high-shear mixing (IHSM) method was used to produce emulsions, and three protein contents were investigated at a fixed RPO content of 12%: low (7.7%), medium (10.5%) and high (13%). Pressure drop measurement was used as an inline approach to determine viscosity using the Hagen-Poiseuille equation. In addition, offline viscometry, particle size and emulsion stability analyses were performed. Emulsion fat droplet size decreased significantly ($P < 0.05$) as a function of number of passes through the IHSM, due to an effective increase in residence time. Furthermore, inline pressure drop data demonstrated that the emulsification process displayed two distinct stages: (i) oil injection, and (ii) reduction in fat droplet size, irrespective of protein content. © 2017 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Applications of hydrodynamic cavitation for instant rehydration of high protein milk powders

Pathania S.; Ho Q.T.; Hogan S.A.; McCarthy N.; Tobin J.T.

Journal of Food Engineering, 2018

The aim of this study was to evaluate the effectiveness of an in-line hydrodynamic cavitation (HC) system, for rehydration of milk protein concentrate powders (MPC) at semi-industrial pilot scale. MPC powder was dispersed in water at 50 °C at 20% (w/w) dry matter (DM) with two commonly used high-shear powder inductors/mixers. The MPC dispersions created were then passed through the HC system to assess subsequent hydration behaviour of the MPC powders. Particle size distribution (PSD) of MPC dispersions prepared using conventional high-shear mixing indicated that complete rehydration of MPC powders was not achieved, with an average D90 and D[4,3] values of 21.17 µm and 5.62 µm respectively, observed in MPC dispersions. In contrast MPC dispersions subjected to HC had a PSD indicative of complete rehydration, with an average D90 and D[4,3] values of 0.45 µm and 0.19 µm, respectively. Apparent viscosity decreased significantly ($p \leq 0.05$) post HC compared to dispersions subjected to conventional high shear mixing. Phase separation profiles showed that HC treated MPC dispersions had increased stability to sedimentation compared to high-shear treated samples. Wetting, immersion, dissolution and solubilisation of high protein powders occurred instantaneously (and simultaneously) during HC. This emerging technology has the potential to achieve complete rehydration of powders in significantly less time than conventional rehydration processes employed by dairy and other industries. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Modelling the changes in viscosity during thermal treatment of milk protein concentrate using kinetic data

Ho Q.T.; Murphy K.M.; Drapala K.P.; Fenelon M.A.; O'Mahony J.A.; Tobin J.T.; McCarthy N.A.

Journal of Food Engineering, 2019

This work aimed to model the effect of heat treatment on viscosity of milk protein concentrate (MPC) using kinetic data. MPC obtained after ultrafiltration was subjected to different time-temperature heat treatment combinations. Heat treatment at high temperature and short time (i.e., 100 or 120 °C×30 s) led to a significant increase in viscosity in MPC systems. Second-order reaction kinetic models proved a better fit than zero- or first-order models when fitted for viscosity response to heat treatment. A distinct deviation in the slope of the Arrhenius plot at 77.9 °C correlated to a significant increase in the rate of viscosity development at temperatures above this, confirming the transition of protein denaturation from the unfolding to the aggregation stage. This study demonstrated that heat-induced viscosity of MPC as a result of protein denaturation/aggregation can be successfully modelled in response to thermal treatment, providing useful new information in predicting the effect of thermal treatment on viscosity of MPC. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Reducing stickiness in spray dried dairy emulsions

O'Neill G.J.; Hollingsworth A.; Harbourne N.; O'Riordan E.D.

Food Hydrocolloids, 2019

High-protein intake in early childhood may cause negative health effects in later life. However, low protein powdered follow-on milks are challenging to manufacture due to stickiness caused by increased free fat and lower glass transition points. Skim milk powder and palm oil emulsions, ranging from 0.25 to 5% w/w protein were assessed for their stability and subsequently spray dried to produce powders ranging from 1 to 20% w/w protein. Powders containing 2–8% w/w protein had high free fat levels despite the initial emulsion being stable, suggesting small adjustments in protein concentration can have a significant impact on emulsion stability during drying and atomisation. Reducing the protein content via substitution with lactose reduced the powders glass transition point (T_g) from 65 to 49 °C (powder contained 4%w/w protein, 67%w/w carbohydrate and 28%w/w fat). Substituting lactose with maltodextrin (DE6) at a 45%w/w level of inclusion increased the T_g of the powders from 50 to 113 °C, however, viscosity increased significantly. Interestingly, using glucose syrup (DE39) to substitute lactose at a 15%w/w level of inclusion, increased the powder T_g by 21 °C with no significant change in emulsion viscosity compared to a control with lactose as the sole carbohydrate. XPS analysis showed fat type (milk, palm, sunflower) influenced powder surface fat coverage. The inner fat contained increased levels of saturated fatty acids (palmitic and stearic) compared to the surface and encapsulated fat. In conclusion the T_g of low protein spray dried emulsions can be increased and the surface fat decreased, helping to reduce susceptibility to stickiness. © 2018

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Funding Source: DPTC 1

Research Programme: Pillar 3

Elucidation of factors responsible for formation of white flecks in reconstituted fat filled milk powders

Schmidmeier C.; O'Gorman C.; Drapala K.P.; Waldron D.S.; O'Mahony J.A.

Colloids and Surfaces A: Physicochemical and Engineering Aspects, 2019

Fat filled milk powders (FFMP) are formulated by blending skim milk and vegetable oil, to which lactose, permeate, sugar, maltodextrin, vitamins and minerals may also be added. The liquid mix is usually homogenised and spray dried, with agglomeration and lecithination, to produce powders for a range of end-user applications including drinking milk, yoghurt base and coffee whitening. Thus, it is important to avoid common powder defects such as inconsistent whitening, feathering or white flecking on reconstitution. The phenomenon of white flecking, and in particular, the underlying causes of fleck formation, are poorly understood. To better understand white fleck formation, six size fraction samples from two different FFMPs, a good (low level flecking) and a poor powder (high level flecking), were profiled. Most extensive flecking was observed in the coarse fraction of the poor powder, which also had an 8-fold higher free fat content than all other fractions and displayed poor emulsion stability ($D_{3,2} = 15 \pm 3.9 \mu\text{m}$ compared to $0.45\text{--}0.75 \mu\text{m}$ for all other fractions). Treatment of the reconstituted emulsions with an anionic surfactant or a reducing agent suggested that integrity of white flecks was based mainly on electrostatic interactions between proteins, with little contribution from covalent bonds. Although the extent of whey protein denaturation ranged between 22.6–47.5%, whey protein insolubility appeared to play a sub-ordinate role in white fleck formation. These results suggest that presence of flecks in FFMP was mainly associated with poor thermal stability of the emulsions. © 2019 Elsevier B.V.

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Funding Source: DPTC 1

Research Programme: Pillar 3

The effect of agglomerate integrity and blending formulation on the mechanical properties of whey protein concentrate powder tablets

Schmidmeier C.; Wen Y.; Drapala K.P.; Dennehy T.; McGuirke A.; Cronin K.; O'Mahony J.A.

Journal of Food Engineering, 2019

In this work, the strength of tablets made from mixtures of whey protein concentrate (WPC) powder and different types of sugar was investigated. The agglomerated WPC powder was pneumatically conveyed (lean phase) at three different air speeds of 10, 20 or 30 m/s to simulate the effects of industrial powder transport, resulting in agglomerates with reduced integrity. Conveyed and control WPC powder, and sugar of three different particle size distributions (i.e., granulated, GS; caster, CS and icing sugar, IS) were analysed for microstructure, particle size, moisture, and flowability. Tensile strength of tablets from WPC powder alone or blended with 0.5–10% sugar was tested. Conveying of WPC powder led to a significant increase in tablet strength, (i.e., from failure strength of 1.67 N/mm² for control tablets to 2.33, 2.11 and 2.11 N/mm² after conveying at 10, 20 or 30 m/s, respectively), suggesting that the generation of smaller powder particles led to increased mechanical strength of tablets. The addition of sugar also resulted in a significant increase in tablet strength (e.g., failure strength of 2.34, 2.12 and 2.47 N/mm² at 1.5% addition level of GS, CS and IS, respectively) and strongest tablets were obtained at 1.5–3% sugar addition and by blending with icing sugar. This study demonstrated, for the first time, that conveying and dry blending of whey protein and sugar powders significantly influenced the rheological properties of resultant tablets. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

A cascade microfiltration and reverse osmosis approach for energy efficient concentration of skim milk

Blais H.; Ho Q.T.; Murphy E.G.; Schroën K.; Tobin J.T.

Journal of Food Engineering, 2021

To improve the efficiency of water removal from skim milk, a cascade membrane process of microfiltration and reverse osmosis (RO) was developed whereby skim milk was concentrated to 18% dry matter (DM) by RO at either 15 or 50 °C. The average flux of the RO process at 50 °C was 89% higher than that observed at 15 °C, linked to altered membrane surface fouling behaviour due to lower viscosity, higher cross-flow velocity and increased diffusivity of the solvent phase. In corollary, a ~57% energy reduction per unit volume of water removed was observed when the RO process was operated at 50 °C. Evaluation of the physicochemical properties of control (9% DM content skim milk) and RO retentates post-heating (at 80, 90 and 120 °C) and post-evaporation (to 42% DM) demonstrated a clear relationship between heating at elevated DM contents and solution viscosity, an effect that was compounded at higher heating temperatures. © 2021 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Cold microfiltration as an enabler of sustainable dairy protein ingredient innovation

France T.C.; Kelly A.L.; Crowley S.V.; O'mahony J.A.

Foods, 2021

Classically, microfiltration (0.1–0.5 μm) of bovine skim milk is performed at warm temperatures (45–55°C), to produce micellar casein and milk-derived whey protein ingredients. Microfiltration at these temperatures is associated with high initial permeate flux and allows for the retention of the casein fraction, resulting in a whey protein fraction of high purity. Increasingly, however, the microfiltration of skim milk and other dairy streams at low temperatures ($\leq 20^\circ\text{C}$) is being used in the dairy industry. The trend towards cold filtration has arisen due to associated benefits of improved microbial quality and reduced fouling, allowing for extended processing times, improved product quality and opportunities for more sustainable processing. Performing microfiltration of skim milk at low temperatures also alters the protein profile and mineral composition of the resulting processing streams, allowing for the generation of new ingredients. However, the use of low processing temperatures is associated with high mechanical energy consumption to compensate for the increased viscosity, and thermal energy consumption for inline cooling, impacting the sustainability of the process. This review will examine the differences between warm and cold microfiltration in terms of membrane performance, partitioning of bovine milk constituents, microbial growth, ingredient innovation and process sustainability. © 2021 by the authors. Licensee MDPI, Basel, Switzerland.

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Funding Source: DPTC 1

Research Programme: Pillar 3

The effects of temperature and transmembrane pressure on protein, calcium and plasmin partitioning during microfiltration of skim milk

France T.C.; Kelly A.L.; Crowley S.V.; O'Mahony J.A.

International Dairy Journal, 2021

Dissociation of β -casein from casein micelles at low temperature is exploited in the manufacture of β -casein-enriched ingredients. In this study, the effects of selected temperatures within the cold processing range (4, 8, 12, 16 and 20 °C) and different transmembrane pressures (0.05 and 0.30 bar), on protein, mineral and plasmin partitioning during microfiltration were determined. Significantly higher ($p < 0.05$) total and ionic calcium concentrations were measured in permeate generated at 4 °C under sub-critical flux conditions (0.05 bar) compared with limiting flux conditions (0.30 bar). Under the former, the highest concentration of β -casein in permeate (2.02 mg mL⁻¹) was achieved at 4 °C; however, these conditions also led to the highest plasmin activity (0.023 AMC units mL⁻¹) in permeate. Lower processing temperature and reduced fouling contribute to higher yields of β -casein, although the concomitantly higher plasmin activity may contribute to protein stability challenges during down-stream processing of such permeates. © 2020 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

The Plasmin System in Milk and Dairy Products

France T.C.; O'Mahony J.A.; Kelly A.L.

Food Engineering Series, 2021

Plasmin is the principal indigenous proteinase present in bovine milk, and has been the focus of many studies as it contributes in diverse ways to the quality of milk and milk-based products. The plasmin system is a complex protease-protease inhibitor system, which involves a series of interactions that ultimately result in the activation of plasminogen to active plasmin. The components of the plasmin system within bovine milk have been successfully quantified, isolated and characterised. Components of the plasmin system are affected by numerous factors such as processing conditions, environmental factors and storage conditions, which ultimately alter the rate/extent of plasminogen activation and plasmin-induced proteolysis. Factors such as pH, heat treatment, presence of whey proteins and temperature of storage can also influence the rate of plasmin-induced hydrolysis of caseins in milk and other dairy products. Plasmin-mediated hydrolysis can have both beneficial and negative effects on a wide variety of dairy products; plasmin is of great importance in the development of flavour and texture during cheese ripening for example, whereas, in ultra-high-temperature milk and high protein dairy-based beverages, plasmin-induced proteolysis can cause undesirable gelation. In this chapter, an overview of the current state of knowledge and areas that require additional research is presented. © 2021, Springer Nature Switzerland AG.

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Funding Source: DPTC 1

Research Programme: Pillar 3

Agents of Change: Enzymes in Milk and Dairy Products

Kelly, A.L.; Bach Larsen, L. (Editors)

Food Engineering Series (Book), 2021

Milk has been converted into dairy products for millennia, firstly perhaps by accident with the discoveries of transformation of milk to curds and whey by animal derived coagulants and the benefits of fermentation by adventitious microorganisms.

In recent decades, the application of scientific study to what, in many cases, were traditional processes has illustrated the roles of enzymes in many different aspects of dairy products, whether deliberate (in the use of rennet to make cheese), natural and desirable (the antimicrobial activities of many enzymes) or natural and undesirable (bacterial spoilage or development of lipolytic rancidity). There is no doubt that milk and dairy products comprise complex enzyme systems, and enzymes truly are agents of change for many dairy products; in the current parlance, one could be tempted to use the 'omics' terminology to refer to the dairy 'enzyme'. These enzymes are typically considered in most textbooks and reference works on dairy science and technology, and there have been conferences dedicated to the topic, such as the First International Conference on Indigenous Milk Enzymes, organised by the International Dairy Federation and held in Cork, Ireland, in April 2005 (which the co-editors of this book were both involved in organising). However, there has been no reference book solely dedicated to this topic, despite all aspects of milk and dairy product enzymology being active current areas of research (e.g., in the area of applications of cross-linking enzyme or the characterisation of psychrotrophic bacterial enzymes). Thus, the objective of this book is to gather in one place a collection of diverse articles on all the elements of this complex element of dairy science, written by experts in their relevant areas, such that the result is a reference which reflects the very state of current knowledge of the field and should be of use for many years to come for those involved in, studying or applying in industry dairy science and processing. As editors, we would like to firstly thank Daniel Falatko at Springer Nature for encouraging and accepting our proposal for this book and to Sofa Valsendur for support and assistance in the production process of the manuscript. We would next like to thank all the authors whose work makes up this book—we were delighted that so many true experts on their own field accepted our invitation to contribute to this project, and thanks to their excellent contributions that this will hopefully be a valuable and unique resource in a very important area of dairy science for many years to come; we also thank our authors for their cooperation and patience during the preparation of this book and their shared enthusiasm for the project, which made our job as editors a pleasure. We also would like to thank Dylan Kelly and, in particular, Ciara Tobin for their local editorial help in manuscript preparation and formatting, Jørgen Gamborg Andersen for fruitful discussions and many colleagues for useful and helpful discussions and suggestions.

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Funding Source: DPTC 1

Research Programme: Pillar 3

The Production of Bioactive Peptides from Milk Proteins

Kleekayai T.; Cermeño M.; FitzGerald R.J.

Food Engineering Series, 2021

Peptides derived from milk proteins are associated with a range of different bioactivities, e.g., antioxidant, antihypertensive, antidiabetic, immunomodulatory, antimicrobial, opioid properties. Peptides can be released following in vitro enzymatic hydrolysis, fermentation and in vivo digestion approaches, alone or in combination. The release of bioactive peptides (BAPs) from milk proteins by these processes is reviewed herein. Furthermore, the contribution of in silico approaches in the targeted release and identification of BAPs is outlined. Details of bioactive milk protein derived peptide sequences obtained by enzymatic hydrolysis, fermentation, and in vivo digestion, as well as by using in silico approaches are presented. Examples of the application of membrane processing and chromatographic techniques for milk BAP fractionation and enrichment are described. Research on the production and identification of milk-derived BAPs can contribute to a better understanding of the nutritional benefits of dairy product consumption. © 2021, Springer Nature Switzerland AG.

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Funding Source: DPTC 1

Research Programme: Pillar 3

The impact of protein standardisation with liquid or powdered milk permeate on the rheological properties of skim milk concentrates

Tsermoula P.; Drapala K.P.; Joyce A.M.; Hoare K.; Crowley S.V.; O'Mahony J.A.

International Dairy Journal, 2021

The objective of this study was to determine the effect of mineral addition during standardisation on colloidal and rheological properties of skim milk on concentration. For that purpose, solubility, ionic strength and ionic calcium activity of milk permeate were determined and skim milk was standardised with either lactose or milk permeate (in powder or reconstituted form). Results indicated that milk permeate contains mineral complexes that alter the ionic equilibrium of skim milk and standardisation with permeate powder has the potential to decrease casein micelle size and integrity. Apparent viscosity of the concentrates demonstrated that skim milk standardised with milk permeate powder had the highest viscosity, followed by that standardised with reconstituted milk permeate or lactose. These novel results demonstrate that the physical state of minerals in milk permeate influence the physicochemical properties of skim milk and thereby affect viscosity of resultant concentrates. © 2021 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Impact of thermal inactivation conditions on the residual proteolytic activity and the viscosity properties of whey protein concentrate enzymatic hydrolysates

Amigo-Benavent M.; FitzGerald R.J.

Food Hydrocolloids, 2022

The rationale for enzymatically hydrolysis of whey proteins includes enhancement of their technofunctional properties and the release of bioactive peptide sequences. This study investigated the impact of thermal treatments, i.e., heating using a water bath, using a tubular heat exchanger and heating during spray drying, on the residual proteolytic activity (RA), apparent viscosity (η_{app}) and gelation temperature (T_g) of whey protein concentrate hydrolysates (WPH) generated with Alcalase® (WPH-Alc), Neutrase™ (WPH-Neu) and Debitrase® (WPH-Deb). The WPHs generated showed different degrees of hydrolysis (DH, 2.60–11.13%) and chromatographic profiles. Thermal inactivation studies showed different impacts on RA dependent on the thermal treatment conditions. The η_{app} of unheated WPHs were significantly lower (16–18%) than that of the unhydrolysed whey protein concentrate (WPC). Thermal inactivation (80 °C waterbath) increased η_{app} of all samples producing aggregates. Tubular heat exchanger inactivation increased η_{app} of WPH-Alc and WPH-Deb at different rates, whereas no significant changes were observed for WPH-Neu for holding times lower than 10 min. At 45% (w/v) total solids (TS) the viscosity of WPC was 1.0 Pa s, whereas the equivalent viscosity for WPC -Alc and WPH-Deb was achieved at 49.5 and 50.1% TS. Different trends were observed in T_g of the WPHs which were dependent on the enzyme and thermal treatment. Overall, this study demonstrates that judicious choice of thermal treatments during WPH enzyme inactivation is required in order to achieve an appropriate balance between RA, η_{app} and T_g . © 2021 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 3

Optimisation of low temperature (8°C) enzymatic hydrolysis of acid whey using design of experiments (DOE) for the generation of thermally stable whey protein hydrolysates

Kleekayai T.; Singh U.; FitzGerald R.J.

Food Hydrocolloids, 2024

The impact of enzymatic hydrolysis at 8°C of liquid whey protein concentrate (WPC35) derived from acid casein manufacture using Prolyve 1000®, a microbial protease preparation, under free-fall pH conditions on the physicochemical and the thermal stability properties of the resultant hydrolysates was investigated. The hydrolysis process was optimised using a design of experiments (DOE) approach. Eleven hydrolysates (H1–H11) were generated using 3 factors x 2 levels, i.e., enzyme:substrate (E:S) 0.25–1.00% (v/w), starting pH 7.5–9.0 and incubation time 10–96 h. Hydrolysate degree of hydrolysis (DH) values ranged from 0.75 (H10) to 4.74% (H6) which increased to 1.91 (H10) and 7.24% (H6) following subsequent thermal processing (mimicking the evaporation process) at 54°C x 15 min and 64°C x 10 min. The apparent viscosity (η_{app}) of all hydrolysates was measured to assess their heat stability during heating at 85°C for up to 20 min, this was shown to be E:S and pH-dependent. While the unhydrolysed samples formed a gel on heating at ~74–80°C. Following DOE analysis, it was found that E:S had a significant impact on the DH, the extent of intact protein degradation (%Deg) and the thermal stability. The %Deg following processing at 64°C showed a strong correlation ($R^2 = 0.924$; $p < 0.001$) with η_{app} , and thus this parameter may be used to predict the thermal stability of the hydrolysate samples. The optimised hydrolysis conditions for the generation of heat stable WPC35 hydrolysates during 8°C incubation were achieved using an E:S = 0.625% at a starting pH = 8.65. Performance of protein hydrolysis reactions at low temperature has potential to enhance process sustainability. © 2023 The Authors

Link to article: <https://doi.org/10.1016/j.foodhyd.2023.109351>

DPTC 1 Publications: Pillar 4

Funding Source: DPTC 1

Research Programme: Pillar 4

Non-invasive 3D and 360° optical imaging of micro-particles

El Arnaout T.; Cullen P.J.

Scientific Reports, 2017

Scanning electron microscopy and X-ray microtomography are useful methods for high resolution shape imaging. Visible microscopy is also common, however, developing a low-cost and customizable system for surface and shape investigation of optically active particles is challenging. In this work, we demonstrate an assembly offering good light sensitivity, flexibility of illumination and contrasts from varying angles. The design was applied, together with recent programs for focus-stacking, to analyze crystals of taurine, L-glutamic acid, acetylsalicylic acid, and copper sulfate, along with digital 3D-360° modelling of phosphorescent [Ru(bpy)₃]Cl₂ and strontium aluminate particles. We further tested the approach for real time monitoring of size, shape and texture analysis of fat filled milk particles and acid whey powders. The findings show proof of concept for detailed feature imaging of particles directly from the process environment. © 2017 The Author(s).

Link to article: <https://doi.org/10.1038/s41598-017-06830-8>

Funding Source: DPTC 1

Research Programme: Pillar 4

Determination of *Listeria monocytogenes* numbers at less than 10 cfu/g

Hunt K.; Vacelet M.; Jordan K.

Irish Journal of Agricultural and Food Research, 2017

Listeria monocytogenes is a foodborne pathogen that causes a relatively rare foodborne disease called listeriosis, with a high mortality rate of 20%-30% and an undefined dose response. Current European Union regulations permit up to 100 colony-forming units (cfu)/g in food at the end of its shelf life, where the food has been shown not to support the growth of this pathogenic bacterium. Therefore, enumeration of *L. monocytogenes* at low numbers in food is important. The objective of this study was to reduce the detection limit of *L. monocytogenes* in food by a factor of 10. The International Organisation for Standardisation (ISO) 11290-2 method for enumeration of *L. monocytogenes* in food recommends spreading 0.1 mL of a 1:10 dilution of the food on the surface of an agar plate (detection limit 100 cfu/g), or 1.0 mL spread in equal parts on the surface of three agar plates (detection limit: 10 cfu/g). The pour-plate method (using 1 or 10 mL of an appropriate dilution) was compared to the spread-plate method using the ISO-approved chromogenic medium Agar *Listeria* according to Ottaviani and Agosti (ALOA). Using the pour-plate method, the colony morphology and halo formation were similar to the spread-plate method from pure cultures and inoculated foods. Using the pourplate method in a 140 mm Petri dish, 10 mL of a 1:10 dilution of food allowed determination of numbers as low as 1 cfu/g. Applying this method, *L. monocytogenes* in naturally contaminated food samples were enumerated at numbers as low as 1-9 cfu/g. © 2017, Teagasc. All rights reserved.

Link to article: <https://doi.org/10.1515/ijafr-2017-0004>

Funding Source: DPTC 1

Research Programme: Pillar 4

Listeria monocytogenes in the Food Processing Environment

Jordan K.; Hunt K.; Lourenco A.; Pennone V.

Current Clinical Microbiology Reports, 2018

Purpose of Review: *Listeria monocytogenes* is a foodborne pathogen that causes listeriosis, a relatively rare but potentially fatal disease with a 19% mortality rate and a 99% hospitalisation rate. It affects mainly elderly and immunocompromised individuals. Ready-to-eat (RTE) foods are particularly dangerous with regard to *L. monocytogenes* as there is no further anti-microbial step between production and consumption. The purpose of this work is to review the importance of *Listeria monocytogenes* in the food processing environment. **Recent Findings:** Cross-contamination from the processing environment to the food at production or at retail level is the most common route of RTE food contamination. If present on a food matrix, *L. monocytogenes* has a remarkable ability to survive and can grow during refrigeration to sufficient numbers to cause disease. **Summary:** While hygiene processes and awareness can help control of *L. monocytogenes* in food processing environments, new methods such as bacteriophages and bacteriocins are being applied to control it in food, reducing public health issues. © 2018, Springer International Publishing AG, part of Springer Nature.

Link to article: <https://doi.org/10.1007/s40588-018-0090-1>

Funding Source: DPTC 1

Research Programme: Pillar 4

Process analytical technology for cheese manufacture

Panikuttira B.; O'Shea N.; Tobin J.T.; Tiwari B.K.; O'Donnell C.P.

International Journal of Food Science and Technology, 2018

Recent research on the development and application of process analytical technology (PAT) for cheese manufacture is reviewed in this article. PAT is a framework for innovative process manufacturing and quality assurance, which has been widely investigated for dairy processing applications, where particular processing challenges arise due to the variations in the physiochemical properties of milk. Cheese manufacturers are increasingly considering the adoption of a PAT approach to facilitate manufacture of cheese with enhanced product quality, safety and process efficiency. However, to date adoption of PAT in the dairy industry has been limited due to challenges associated with development and validation of calibration models, instrument variability, sanitary design and compatibility with processing environments. New technical developments in PAT tools, advances in chemometric modelling, robust data management tools and improved understanding of critical product and process parameters will facilitate further adoption of a PAT approach in cheese manufacture. © 2018 Institute of Food Science and Technology

Link to article: <https://doi.org/10.1111/ijfs.13806>

Funding Source: DPTC 1

Research Programme: Pillar 4

Flow cytometry as a potential method of measuring bacterial viability in probiotic products: A review

Wilkinson M.G.

Trends in Food Science and Technology, 2018

- A review of flow cytometry for monitoring [probiotic](#) culture viability was undertaken.
- Flow cytometry had good correlation with [plate counts](#) for concentrated fresh cultures.
- Sample clean up steps are extremely important to achieve optimal resolution.
- Flow cytometry generates multi-parametric data not always reflecting plate counts.
- Regulatory definitions of [cell viability](#) and cell vitality needs clarification.

Link to article: <https://doi.org/10.1016/j.tifs.2018.05.006>

Funding Source: DPTC 1

Research Programme: Pillar 4

Evaluation and validation of an inline Coriolis flowmeter to measure dynamic viscosity during laboratory and pilot-scale food processing

Bista A.; Hogan S.A.; O'Donnell C.P.; Tobin J.T.; O'Shea N.

Innovative Food Science and Emerging Technologies, 2019

Inline process analytical technology tools are increasingly employed in industry as they facilitate real time process parameter monitoring and optimization of product quality. Use of an inline viscometer in milk powder manufacture allows greater process control by monitoring the dynamic viscosity of the process concentrate pumped to the spray drier. In this study, an inline Promass I300 was used to measure the dynamic viscosity of both a Newtonian fluid (10–60% w/w sucrose solutions) and a non-Newtonian fluid (10–40% w/w skim milk concentrate) at laboratory scale at 25 °C. Validation of the instrument for measurement of a non-Newtonian fluid was completed at pilot-scale under similar operating conditions. Coefficient of determination ($R^2 = 0.99$) was obtained between inline and offline viscosity measurements for both Newtonian and non-Newtonian fluids investigated. This study demonstrated the potential of an inline Coriolis flowmeter for rapid and accurate measurement of dynamic viscosity during processing of dairy streams. Industrial relevance: Control of milk concentrate viscosity during the manufacture of milk powder is critical to reduce energy consumption, reduce fouling and meet powder functional properties e.g. dispersability, flowability, particle size and powder hydration. The use of inline instruments for continuous monitoring of viscosity offers economic benefits to the producer as it can contribute to reduction in commonly encountered process issues such as fouling, blocking of nozzles and evaporator, which can result in process downtime and in extreme cases, product rework. Inline instruments can thus be employed to improve process control and reduce production waste during the manufacture of dairy powders. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.ifset.2019.05.004>

Funding Source: DPTC 1

Research Programme: Pillar 4

Occurrence and identification of spore-forming bacteria in skim-milk powders

Li F.; Hunt K.; Van Hoorde K.; Butler F.; Jordan K.; Tobin J.T.

International Dairy Journal, 2019

The different customer and regulatory specifications for mesophilic and thermophilic aerobic and anaerobic spore numbers in skim-milk powder, in addition to some specifications on specific spore-forming bacteria, such as *Bacillus cereus*, can be challenging for the industry to meet. Twenty-two samples of medium-heat skim-milk spray-dried powder from eight sources were analysed in triplicate with 16 bacterial and spore enumeration tests to understand the variety of spore-forming bacteria population. Using 16S rDNA sequencing, the species were identified for 269 isolates that were representative of the various tests. Of the isolates identified, 68% were *Bacillus licheniformis*, a facultative anaerobe that can survive and grow at mesophilic and thermophilic temperatures, making it difficult to eliminate in manufacturing environments. Using whole genome sequencing, 16 of 23 isolates identified as *B. licheniformis* by 16S sequencing were confirmed as *B. licheniformis*, four were identified as *Bacillus paralicheniformis* and three were identified as *Bacillus* sp. H15-1. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.idairyj.2019.05.004>

Funding Source: DPTC 1

Research Programme: Pillar 4

The application of process analytical technologies (PAT) to the dairy industry for real time product characterization - process viscometry

O'Shea N.; O'Callaghan T.F.; Tobin J.T.

Innovative Food Science and Emerging Technologies, 2019

The ideal PAT tool is an inline instrument that can monitor and measure process parameters simultaneously in real time while operating in a highly automated environment. Instruments must be of sanitary design, operate robustly within the full process cycle (production and cleaning). Inline determination of the rheological properties of moving fluids (i.e. dairy concentrates) is one of the process parameters where PAT tools can add real value in terms of optimising process control. Measurement of process viscosity is crucial in the monitoring and control of a variety of concentration processes in the dairy industry. Continuous monitoring of the rheological behaviour of the fluid can allow for optimisation of the process e.g. pumping (avoid pump blockage and failure), evaporation (limit fouling and maximise water removal) and spray drying (avoidance of nozzle fouling). This review concentrates on the state of the art developments being made in the area of process viscometry. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.ifset.2019.05.003>

Funding Source: DPTC 1

Research Programme: Pillar 4

Evaluation of a fluorescence and infrared backscatter sensor to monitor acid induced coagulation of skim milk

Panikuttira B.; Payne F.A.; O'Shea N.; Tobin J.T.; O'Donnell C.P.

Innovative Food Science and Emerging Technologies, 2019

A prototype sensor that employs both ultraviolet excited fluorescence and infrared light backscatter was evaluated as an in-line process analytical technology (PAT) tool to monitor acid induced coagulation kinetics of skim milk. Coagulation experiments were carried out at 32 °C using three concentrations of glucono-delta-lactone (GDL). Measurement of storage modulus (G') of acidified skim milk gel was used as a reference rheological method to monitor the coagulation kinetics. Prediction models were developed to predict the times required for acidified skim milk coagulum to reach selected G' values (0.5 Pa, 1 Pa, 5 Pa, 10 Pa and 15 Pa) using time parameters extracted from the ultraviolet excited fluorescence and infrared light backscatter profiles. A strong correlation was observed between the predicted times developed using time parameters extracted from the prototype sensor profiles and the measured G' times extracted from the rheometer ($R^2 = 0.97$, standard error of prediction = 2.8 min). This study concluded that the prototype fluorescence and infrared backscatter sensor investigated combined with the developed rheological prediction model can be used as a potential PAT tool for in-line monitoring of coagulation kinetics in the manufacture of acid induced milk gels. Industrial relevance: The prototype fluorescence and infrared backscatter sensor investigated in this study combined with the developed rheological prediction model can be employed to monitor and control coagulation kinetics in a wide range of dairy processing applications including fresh cheese varieties and yoghurt manufacture. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.ifset.2019.04.011>

Funding Source: DPTC 1

Research Programme: Pillar 4

Monitoring Viscosity and Total Solids Content of Milk Protein Concentrate Using an Inline Acoustic Flowmeter at Laboratory Scale

Bista, A; Tobin, JT; O'Donnell, CP; O'Shea, N

Foods, 2020

Control of milk concentrate viscosity and total solids (TS) content prior to spray drying can improve dairy ingredient manufacture. However, the availability of hygienic and appropriately pressure rated process viscometers for inline monitoring of viscosity is limited. An acoustic flowmeter (FLOWave) is an inline process analytical technology (PAT) tool that measures changes in acoustic signals in response to changes in liquid properties (i.e., acoustic transmission (AT), acoustic impedance (AI), temperature and volume flowrate). In this study, an acoustic flowmeter is evaluated as an inline PAT tool for monitoring viscosity of milk protein concentrate (MPC85), protein and TS content of (MPC85), and standardised MPC (sMPC). Laboratory scale experiments were carried out at 45 °C for five different concentrations (4–21%) of MPC85 and sMPC. Results showed that AT decreased with an increase in MPC85 viscosity (e.g., AT was $98.79 \pm 0.04\%$ and $86.65 \pm 0.17\%$ for 4% and 21% TS content, respectively). Non-linear regression was carried out to develop a relationship between AT and offline viscosity (R^2 (coefficient of determination) value = 0.97 and standard error of prediction = 1.86 mPa·s). AI was observed to increase at higher protein and TS content which was dependent on protein to total solid ratio (P_TSR). Multiple linear regression was carried out to develop the relationship between AI, protein content, TS content and P_TSR. Results demonstrated that AI could be used to monitor the protein and TS content of milk protein concentrate ($R^2 > 0.96$). Overall this study demonstrated the potential of an inline acoustic flowmeter for monitoring process viscosity, protein and TS during dairy concentrate processing.

Link to article: <http://dx.doi.org/10.3390/foods9091310>

Funding Source: DPTC 1

Research Programme: Pillar 4

Investigation of an in-line prototype fluorescence and infrared backscatter sensor to monitor rennet-induced coagulation of skim milk at different protein concentrations

Panikuttira B.; Payne F.A.; O'Shea N.; Tobin J.T.; O'Callaghan D.J.; O'Donnell C.P.

International Journal of Food Science and Technology, 2020

Coagulation of milk is one of the most important steps in cheese manufacture. Cutting the coagulum at optimum firmness is important to optimise the yield and quality of the cheese produced. The aim of this study was to investigate a prototype sensor to monitor rennet-induced coagulation of skim milk at different protein concentrations (3.3%, 4.0% and 4.7%) and to develop a model to predict the coagulum cutting time at a desired storage modulus (G'). Fluorescence and infrared backscatter profiles were recorded at wavelengths of 350 and 880 nm, respectively. Rheological measurements were used as a reference method to determine the times required for the coagulum to reach G' values of 0.5, 5 and 20 Pa. Time parameters extracted from the optical profiles generated during the coagulation process were used to develop a model to predict the cutting time at which the coagulum reaches selected G' values. This study demonstrated that the investigated prototype sensor, combined with the developed prediction model, can be used as an in-line PAT tool for real-time monitoring of milk coagulation and prediction of cutting time in cheese manufacturing. © 2019 Institute of Food Science and Technology

Link to article: <https://doi.org/10.1111/ijfs.14267>

Funding Source: DPTC 1

Research Programme: Pillar 4

Assessment of a solid-state bulk acoustic wave sensor to measure viscosity of Newtonian and Non-Newtonian fluids under static and flow conditions

Pu Y.; O'Shea N.; Hogan S.A.; Tobin J.T.

Journal of Food Engineering, 2020

Monitoring and control of inline viscosity is crucial for process optimisation and for ensuring a high quality final product but currently this parameter is still under-utilised in the dairy industry. This study investigated a solid-state bulk acoustic wave sensor to measure the viscosity of Newtonian (oil standards) and Non-Newtonian fluids (reconstituted skim milk (RSM) at different concentrations) under static (off-line measurements) and flow conditions (in-line measurements). Results illustrated that an increase in total solids (TS) of RSM gave an increase in acoustic viscosity. Non-linear regression was applied to the experimental data to successfully transform the acoustic viscosity outputs into commonly-used reference viscosity values. RSM at higher TS presented a non-Newtonian behaviour and demonstrated shear-thinning properties. Under flow conditions the viscosity of the RSM decreased as a result of shearing experienced in the pipe. This study demonstrated the potential of an acoustic wave sensor to measure in-line viscosity in dairy applications. © 2020 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.jfoodeng.2020.109917>

Funding Source: DPTC 1

Research Programme: Pillar 4

Review of near-infrared spectroscopy as a process analytical technology for real-time product monitoring in dairy processing

Pu Y.-Y.; O'Donnell C.; Tobin J.T.; O'Shea N.

International Dairy Journal, 2020

Real-time process/product monitoring can be achieved using suitable process analytical technologies (PAT) to improve process efficiencies and product quality. In the dairy industry, near infrared (NIR) spectroscopy has been utilised as a laboratory analytical method (off-line) for compositional analysis of dairy products since the 1970s. Recent advances in NIR technology and instrumentation have widened its applications from a bench-top analytical instrument to a promising PAT tool for on-line and in-line implementation. This review focuses on the use of NIR technology for real-time monitoring of dairy products, by briefly outlining the measurement principle, NIR instrument configurations, in-line sampling methods, calibration models development, some practical considerations for process installation, and current state of the art in on-line and in-line NIR applications (2012 to date) for continuous process monitoring in the production of dairy products. The challenges and additional resources required to improve production efficiencies using NIR spectroscopy are also discussed. © 2019 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 4

A Bayesian estimation of the concentration of microbial organisms in powdered foods arising from repeat testing for microbial contamination

von Westerholt F.; Butler F.

Microbial Risk Analysis, 2020

Microbial food safety sampling in the food industry is generally based on single sampling schemes, with the results dictating the acceptance/rejection of the entire food lot. Resampling plans are generally only used to discern quality of ingredients or food items where food safety is not part of the evaluation. This paper developed a Bayesian model to quantify the uncertainty in the concentration of a bacterial organism in a batch of product considering the results from the primary sampling, as well as from subsequent resampling. Bayesian consideration of the primary sampling outcomes allowed formulation of a prior distribution which was then used to revise the uncertainty in the concentration present. Uncertainty distributions for microbial concentration were calculated for a range of resampling scenarios, in the event where one sample from an initial sampling tested positive. The study demonstrated that even in the event of large numbers of negative retest results, there was only a small shift in the uncertainty distributions of the concentrations of microorganisms present compared to the results from the initial sampling results. When the second set of testing includes positive outcomes, the peak of the uncertainty distribution moves to the right, as the retest outcomes revise upwards the initial estimate of the concentration of the microbial organism present. The study demonstrated the potential value of additional sampling to better estimate the likely microorganism concentration present in the food product. Especially in the events where a large proportion of the retests were negative, the magnitude of uncertainty was improved. The approach may be especially valuable for any investigation into a wider root cause analysis undertaken after the original positive test. © 2019

Link to article: <https://doi.org/10.1016/j.mran.2019.07.004>

DPTC 1 Publications: Pillar 5

Funding Source: DPTC 1

Research Programme: Pillar 5

A review of environmental life cycle assessment studies examining cheese production

Finnegan W.; Yan M.; Holden N.M.; Goggins J.

International Journal of Life Cycle Assessment, 2018

Purpose: Cheese is one of the world's most widely consumed dairy products and its popularity is ever growing. However, as concerns for the environmental impact of industries increase, products like cheese, which have a significant environmental impact, may lose their popularity. A commonly used technique to assess the environmental impact of a product is life cycle assessment (LCA). In this paper, a state-of-the-art review of LCA studies on the environmental impact of cheese production is presented. **Methods:** Sixteen LCA studies, which explored the impact from the production of a variety of cheese types (fresh, mature and semi-hard) were examined and discussed. The four stages of the LCA were examined and the range of results of selected environmental impact categories (global warming potential, acidification potential and eutrophication potential) were detailed and discussed. **Results and discussion:** For each of these environmental impact categories, raw milk production was consistently found to be the most significant contributor to the total impact, which was followed by processing. It was found that allocation between cheese and its by-products was crucial in determining the impact of cheese production and standardisation or guidelines may be needed. Very little information relating to wastewater treatment system and processes were reported and this leads to inaccurate environmental impact modelling relating to these aspects of the manufacture of cheese. Very few studies included the design of packaging in terms of reducing food waste, which may significantly contribute to the overall environmental impact. **Conclusions:** As raw milk production was found to have the greatest contribution to environmental impact, mitigation strategies at farm-level, particularly in relation to enteric fermentation and manure management, need to be implemented. Additionally, based on the literature, there is a suggestion that fresh cheese has less of an environmental impact than semi-hard cheeses, particularly when examining direct energy consumption. However, there needs to be more case studies investigated to justify this statement. © 2017, Springer-Verlag GmbH Germany.

Link to article: <https://doi.org/10.1007/s11367-017-1407-7>

Funding Source: DPTC 1

Research Programme: Pillar 5

Fly Ash Characterization from *Cynara cardunculus* L. Gasification

Serrano D.; Kwapinska M.; Sánchez-Delgado S.; Leahy J.J.

Energy and Fuels, 2018

This study analyzes the characteristics of fines produced during the air-blown gasification of *Cynara cardunculus* L. in a bubbling fluidized bed. These fines are collected by means of two hot cyclones and a hot filter. The gasification temperature is varied from 700 to 800 °C using olivine and magnesite as bed materials, with an equivalence ratio of 0.2. Relatively high carbon content is found in the entrained fines for all experiments. The lower heating value of the elutriated fines varies from 5.2 to 9.4 MJ/kgdb. Around 75% of the fines are captured in the first cyclone, 5% in the second cyclone, and the remaining 20% in the hot filter. The concentration of elements such as Se and Cl makes these fly ashes a hazardous material. Based on these properties the potential reuse of fly ashes is evaluated. © 2018 American Chemical Society.

Link to article: <https://doi.org/10.1021/acs.energyfuels.7b04050>

Funding Source: DPTC 1

Research Programme: Pillar 5

Life cycle assessment of multi-product dairy processing using Irish butter and milk powders as an example

Yan M.; Holden N.M.

Journal of Cleaner Production, 2018

The Irish dairy industry faces a challenging market and a focus on the environmental impacts of products, both of which affect sustainable growth. The objectives of the study were to use life cycle assessment to analyse three products from four companies, to find the major contributors to energy use and greenhouse gas emissions, to understand the variation of environmental impacts and to identify the scope for improvement. Cumulative energy demand (CED) and carbon footprint (CF) of butter, skimmed milk powder (SMP), and fat filled powder (FFP) were calculated. The system boundary was from farm gate to processor gate to facilitate benchmarking. Data quality was generally sufficient, but data gaps were identified for steam sub-metering. Butter CED varied from 6.93 to 9.73 MJ/kg solids, butter CF from 0.41 to 0.62 kg CO₂ eq/kg solids, SMP CED from 24.57 to 27.53 MJ/kg, SMP CF from 1.40 to 1.70 kg CO₂ eq/kg solids, FFP CED was 26.14 MJ/kg solids and FFP CF was 1.65 CO₂ eq/kg solids. Site specific data allowed explanation of variations by differentiating between practices and operational efficiency. Trade-off between renewable energy and ingredients revealed insights to the CF. Reactive power and wastewater treatment capacity demand further research. Valuable recommendations on how to better conduct LCA with industry were provided. This is the first such detailed analysis for the Irish dairy industry. It is important for the industry to move away from average data for impact management and to use site-specific data where possible. The allocation methodology, cascade structure of modelling, survey template, knowledge gaps identified, and the recommendations on conducting LCA with industry contributed to a general framework of LCA of dairy processing. © 2018 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 5

Dairy industry derived wastewater treatment sludge: Generation, type and characterization of nutrients and metals for agricultural reuse

Ashekuzzaman S.M.; Forrestal P.; Richards K.; Fenton O.

Journal of Cleaner Production, 2019

Globally, the processing of milk and dairy products leads to huge volumes of dairy processing wastewater treated sludge. Currently, there are knowledge gaps relating to the volumes generated and the nutrient and metal contents of these sludges. This lack of knowledge prevents the dairy processing industry from making informed pre and post waste generation decisions. Using Ireland as a case study volumes generated are estimated (2012–2017) and a two year seasonal database (2016–2018) across four sludge types (bio-chemically treated activated sludge; lime treated dissolved air flotation processing sludge; a combined treatment sludge and anaerobically digested sludge) utilising samples from nine dairy processing plants was created. Results show that dairy processing sludge increased by 39% in the period up to 126,718 tonnes (wet weight). Database results showed that nutrient contents did not vary seasonally but varied significantly across sludge type and processing plants. The median values (g kg⁻¹ dry weight) for N:P:K for the four sludge types were N: 57.2, 19.5, 46 and 70.4, P: 36.8, 65.9, 20 and 14.6, and K: 7.2, 3.9, 2.9 and 6.1, respectively. Heavy metal concentrations across all samples were significantly lower than those regulated by the European Union for controlling metals accumulation in agricultural land due to sludge recycling. The characterization profile presented in this paper serves as a national and international reference database for future investigations that focus on the valorisation of dairy processing sludge. © 2019 Elsevier Ltd

Link to article: <https://doi.org/10.1016/j.jclepro.2019.05.025>

Funding Source: DPTC 1

Research Programme: Pillar 5

Potential loss of nutrients, carbon and metals in simulated runoff associated with dairy processing sludge application

Ashekuzzaman S.M.; Forrestal P.; Richards K.; Fenton O.

International Journal of Environmental Science and Technology, 2020

Dairy processing wastewater treatment sludge (DPWTS) is applied to grassland to recycle valuable nutrients and organic matter. Until recently the chemical composition of the dominant DPWTS types (calcium, aluminium or iron rich) was unknown, but the latest study (Ashekuzzaman et al. in J Clean Prod 230:1266–1275, 2019) indicates that DPWTS has high nitrogen (N) and phosphorus (P) content and their inherent chemical properties may influence their availability and losses. Herein for the first time edge of field-losses and plant-available P changes are simulated in an agitator test 48 h after application. In terms of overall losses, metals are not of concern across treatment types, iron-rich DPWTS has the highest losses in terms of concentration of ammonium-N and carbon, whereas calcium-rich DPWTS has the highest dissolved reactive P (DRP) losses but lowest losses of ammonium-N and carbon. The loss of DRP was found to be strongly positively associated with the increase in water-extractable P, degree of P saturation, Morgan's and Mehlich-3 extractable phosphorus (P_m, M3-P), respectively. Runoff concentrations presented here represent edge of field-losses and may not represent final concentrations entering a surface waterbody. In terms of plant-available P, there were no negative effects with P_m exhibiting a positive trend across DPWTS types and soils. In particular, a significant increase was found for the calcium-rich DPWTS across all soils (average increase in P_m by fivefold). As applying DPWTS to grassland is an understudied area, further field trials should focus on emissions to air and water and examine the fertilizer value from N and P perspectives. Thus a better management of DPWTS as fertiliser can be maintained from an agronomic and environmental context. © 2020, Islamic Azad University (IAU).

Link to article: <https://doi.org/10.1007/s13762-020-02768-z>

Funding Source: DPTC 1

Research Programme: Pillar 5

Novel Use of Dairy Processing Sludge Derived Pyrogenic Char (DPS-PC) to Remove Phosphorus in Discharge Effluents

Ashekuzzaman S.M.; Kwapinska M.; Leahy J.J.; Richards K.; Fenton O.

Waste and Biomass Valorization, 2020

Purpose: Pyrogenic char (PC) materials derived from the pyrolysis of dairy processing sludge (DPS) could be a cost effective option to develop carbonaceous adsorbent for phosphorus (P) removal from wastewater. The main objectives of the present work were to: (1) determine the efficacy of DPS derived PC (DPS-PC) to remove P from synthetic and dairy wastewater samples, (2) identify possible P removal mechanisms, and identify parameters that could be used to quickly identify the P removal capacity of a char and (3) propose a ranking system for the selection of DPS-PC which includes energy, char yield and P removal criterion. **Methods:** DPS-PC samples were obtained from the pyrolysis process (700 °C) of two sludge streams: (1) bio-chemically treated mixed sludge and (2) lime treated dissolved air floatation (DAF) sludge. Herein, 12 DPS-PC samples were assessed and pre-screened in batch experiments to determine the P removal efficacy from both synthetic and dairy wastewater solutions. The effect of solid to liquid dosage, contact time, pH and P concentration was investigated. Statistical regression and correlation analyses were performed to understand P removal mechanism. The quantitative assessment of char yield, energy balance and P removal performance were combined to propose a ranking system for DPS feedstock selection. **Results:** P removal varied across DPS-PC type and composition, with mixed sludge derived char exhibiting 85–98% P removal at a dose of between 10 and 50 g/L, whereas, those from DAF sludge removed > 99% at 3 g/L. The P removal process was associated with a number of strongly significant mineral phase correlations pertaining to mineral composition (i.e. availability of Ca, Mg and Si) of the DPS-PC samples. A quick water extractable P test together with knowledge of the major P locking minerals can be used to pre-screen the potential of PC for P removal application. This study also provides a physicochemical reference and ranking of DPS feedstock selection, which will be useful for future investigation on the pyrolysis of DPS at pilot-scale and subsequently, to develop PC based efficient adsorbent for application in wastewater treatment. **Graphic Abstract:** [Figure not available: see fulltext.]. © 2019, Springer Nature B.V.

Link to article: <http://dx.doi.org/10.1007/s12649-019-00731-9>

Funding Source: DPTC 1

Research Programme: Pillar 5

Pilot Scale Pyrolysis of Activated Sludge Waste from Milk Processing Factory

Kwapinska M.; Horvat A.; Liu Y.; Leahy J.J.

Waste and Biomass Valorization, 2020

The majority of the sludge from the treatment of wastewater in milk processing plants is land spread. The drawbacks of land spreading include local oversupply due to high transport costs, which results in sludge being spread on lands in the vicinity of the dairy factories. Local oversupply can lead to accumulation of certain substances in soil through annual application over many years. Therefore, in the long term, there is a need for alternative methods to recover energy and nutrients from increasing volumes of sludge generated from dairy processing. Pyrolysis offers a potential alternative to land spreading, which can reduce health and environmental risks, while providing an avenue for the recovery of energy and nutrients. Pyrolysis allows energy recovery in the form of a high calorific value pyrolysis gas and a char which may be used as a soil amendment. In this study pyrolysis of dried dairy sludge was carried out at pilot scale. The results indicate that a dried biological sludge can be successfully pyrolysed and when mixed with wood the resulting char meets European Biochar Certificate criteria regarding carbon content. Most of the initial energy content of the feedstock was retained in the pyrolysis gas prior to cleaning, 53%, compared to 34.5% in the char and 1.5% in the tar. For the pyrolysis gas after cleaning (mainly cracking in presence of air) the initial energy content of the feedstock retained in the gas was only slightly higher than that retained in the char, 39.2% versus 34.5%, while the tar accounted for 0.8% of the initial energy content. © 2019, Springer Nature B.V.

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Research Programme: Pillar 5

Reactor configuration influences microbial community structure during high-rate, low-temperature anaerobic treatment of dairy wastewater

McAteer P.G.; Christine Trego A.; Thorn C.; Mahony T.; Abram F.; O'Flaherty V.

Bioresource Technology, 2020

Low temperature anaerobic digestion remains in its infancy, despite increasing interest for the treatment of complex wastewaters. In this study, the feasibility of low-temperature anaerobic treatment of dairy wastewater was assessed during a 443-day laboratory-scale bioreactor trial. The bioreactors were operated in triplicate at organic loading rates of 7.5–9 kgCODm⁻³d⁻¹ throughout five operational phases. The structure of the microbial community was analysed using quantitative real-time PCR and amplicon sequencing of 16S rRNA genes from DNA and rRNA. The results indicated that low-temperature treatment of dairy wastewater is feasible at 15 °C, but that reactor configuration remains extremely important. The upflow anaerobic sludge bed (UASB) configuration out-performed the expanded granular sludge bed (EGSB)-based configurations. Decreased temperatures resulted in significant reductions in microbiome diversity. Methanosaeta was identified as a dominant genus throughout the trial, while Lactococcus was identified as an important bacterial genus at low-temperatures. However, the relative abundance of Lactococcus was significantly influenced by reactor configuration. © 2020 The Author(s)

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Research Programme: Pillar 5

Grassland Phosphorus and Nitrogen Fertiliser Replacement value of Dairy Processing Dewatered Sludge

Ashekuzzaman S.M.; Forrestal P.; Richards K.G.; Daly K.; Fenton O.

Sustainable Production and Consumption, 2021

Dairy processing sludge is currently a bio-based fertiliser being spread to grassland without knowledge pertaining to its phosphorus (P) or nitrogen (N) fertiliser replacement value. This creates uncertainty of desired crop yield achievement and unproductive nutrient recycling and also poses a great challenge to the dairy milk processing industry in promoting their food processing by-product as valuable recyclable fertiliser. Therefore four representative samples, i.e. two activated sludge (aluminium-precipitated (Al-sludge) and iron-precipitated (Fe-sludge)), and two lime-stabilised calcium-precipitated sludge (Ca1- and Ca2-sludge), were examined at field scale to assess P and N availability for crop yield and uptake in comparison to reference mineral fertilisers over one seasonal year. The field plots were set-up on a light textured clay loam soil within the optimum plant available P (Morgan's soil P index 3, i.e. medium / adequate soil P level) in two separate adjoining areas consisting of P and N availability experiments. Each experiment consisted of 40 plots (each 8×2 m²) of 10 treatments with 4 replications arranged in a randomised complete block design. All dairy sludge (40 kg-P ha⁻¹) and mineral P treatments (rates 0–50 kg-P ha⁻¹) produced similar yields and uptake, and crop P was not affected by sludge applications despite the presence of high Al, Ca and Fe. During the experiment there was no significant change in P index (stayed at index 3) indicating that no treatment caused a decline in P into index 2 (i.e. low soil P level), therefore replacing P removed by the crop. The only change in Morgan's P was observed in the Ca-sludge treatments, but this was due to Morgan's reagent overestimating plant available P in high Ca conditions. From N trial plots a significantly higher grass yield and N uptake was observed for Fe and both Ca-type sludge applied plots than the control (zero N) plot during the 1st harvest, while no statistical difference observed in the subsequent harvests (up to 4th harvesting). The N fertiliser replacement value (derived from mineral N response) of sludge samples was observed to be in the order of Fe (54%)>Ca2 (25%)>Ca1 (22%)>Al (8%) with greater promise of N fertiliser efficiency of Fe and Ca types. Overall these bio-based sludges show promise in recycling P and N for grassland application but longer term trials in other soil types considering other environmental aspects (losses to soil, water and air) can further optimize the management of dairy sludge as an alternative to chemical fertiliser.

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Research Programme: Pillar 5

Solid phase adsorption method for tar sampling – How post sampling treatment affects tar yields and volatile tar compounds?

Horvat A.; Kwapinska M.; Abdel Karim Aramouni N.; Leahy J.J.

Fuel, 2021

The Solid Phase Adsorption (SPA) methodology is the method of choice for sampling tar from biomass gasifiers and pyrolyzers. Reliable and robust tar measurement techniques are essential for having an efficient implementation of these thermochemical processes. This paper critically evaluates the SPA tar sampling method from the following perspectives: (i) the efficiency of tar extraction from the aminopropyl silica sorbent; (ii) the effect of transport and storage period on tar adsorbed on the SPA sorbents; (iii) off-line SPA vs. the on-line μ GC method for quantitative determination of benzene and toluene. The present study showed that the tar extraction efficiency for aminopropyl silica sorbent SPA cartridges was typically 95% or higher. The 30% of tar was lost from these sorbent materials during the airfreight and 21.9% during 5 days of ambient storage, respectively. When two sorbents were employed into the SPA sampling device, the loss was reduced to 15%. The loss of SPA detectable tar was 16.4% when aminopropyl silica sorbents were stored in a freezer for 5 days prior to analysis. A significant portion of light tar compounds (benzene and toluene) was lost from the aminopropyl silica sorbent during the airfreight, while, under either ambient or frozen storage, these compounds were preserved relatively well. The best performance using the SPA method was achieved when tar was extracted from the sorbent immediately on-site. The study also demonstrated the superiority of on-line μ GC measurement over the off-line SPA method for quantitative determination of light tar compounds. Light tar sampling efficiency, such as for benzene and toluene, was improved by introducing the second activated charcoal sorbent. Benzene yields measured by on-line μ GC were up to 10.4 times higher than those extracted from the SPA aminopropyl silica sorbent solely, and, even when two sorbents were employed, they were up to 3.4 times higher. Regarding toluene yields, these values were 2.6 and 1.7 times higher. © 2020 Elsevier Ltd

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Funding Source: DPTC 1

Research Programme: Pillar 5

First proof of concept for full-scale, direct, low-temperature anaerobic treatment of municipal wastewater

Trego A.C.; Conall Holohan B.; Keating C.; Graham A.; O'Connor S.; Gerardo M.; Hughes D.; Ijaz U.Z.; O'Flaherty V.

Bioresource Technology, 2021

Municipal wastewater constitutes the largest fraction of wastewater, and yet treatment processes are largely removal-based. High-rate anaerobic digestion (AD) has revolutionised the sustainability of industrial wastewater treatment and could additionally provide an alternative for municipal wastewater. While AD of dilute municipal wastewater is common in tropical regions, the low temperatures of temperate climates has resulted in slow uptake. Here, we demonstrate for the first time, direct, high-rate, low-temperature AD of low-strength municipal wastewater at full-scale. An 88 m³ hybrid reactor was installed at the municipal wastewater treatment plant in Builth Wells, UK and operated for 290 days. Ambient temperatures ranged from 2 to 18 °C, but remained below 15 °C for > 100 days. Influent BOD fluctuated between 2 and 200 mg L⁻¹. However, BOD removal often reached > 85%. 16S rRNA amplicon sequencing of DNA from the biomass revealed a highly adaptable core microbiome. These findings could provide the basis for the next-generation of municipal wastewater treatment. © 2021

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Funding Source: DPTC 1

Research Programme: Pillar 5

Combined Stochastic and Deterministic Processes Drive Community Assembly of Anaerobic Microbiomes During Granule Flotation

Trego A.C.; McAteer P.G.; Nzeteu C.; Mahony T.; Abram F.; Ijaz U.Z.; O’Flaherty V.

Frontiers in Microbiology, 2021

Advances in null-model approaches have resulted in a deeper understanding of community assembly mechanisms for a variety of complex microbiomes. One under-explored application is assembly of communities from the built-environment, especially during process disturbances. Anaerobic digestion for biological wastewater treatment is often underpinned by retaining millions of active granular biofilm aggregates. Flotation of granules is a major problem, resulting in process failure. Anaerobic aggregates were sampled from three identical bioreactors treating dairy wastewater. Microbiome structure was analysed using qPCR and 16S rRNA gene amplicon sequencing from DNA and cDNA. A comprehensive null-model approach quantified assembly mechanisms of floating and settled communities. Significant differences in diversity were observed between floating and settled granules, in particular, we highlight the changing abundances of *Methanosaeta* and *Lactococcus*. Both stochastic and deterministic processes were important for community assembly. Homogeneous selection was the primary mechanism for all categories, but dispersal processes also contributed. The lottery model was used to identify clade-level competition driving community assembly. Lottery “winners” were identified with different winners between floating and settled groups. Some groups changed their winner status when flotation occurred. Spirochaetaceae, for example, was only a winner in settled biomass (cDNA-level) and lost its winner status during flotation. Alternatively, *Arcobacter butzerli* gained winner status during flotation. This analysis provides a deeper understanding of changes that occur during process instabilities and identified groups which may be washed out—an important consideration for process control. © Copyright © 2021 Trego, McAteer, Nzeteu, Mahony, Abram, Ijaz and O’Flaherty.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Principles, Advances, and Perspectives of Anaerobic Digestion of Lipids

Holohan B.C.; Duarte M.S.; Szabo-Corbacho M.A.; Cavaleiro A.J.; Salvador A.F.; Pereira M.A.; Ziels R.M.; Frijters C.T.M.J.; Pacheco-Ruiz S.; Carballa M.; Sousa D.Z.; Stams A.J.M.; O'Flaherty V.; Van Lier J.B.; Alves M.M.

Environmental Science and Technology, 2022

Several problems associated with the presence of lipids in wastewater treatment plants are usually overcome by removing them ahead of the biological treatment. However, because of their high energy content, waste lipids are interesting yet challenging pollutants in anaerobic wastewater treatment and codigestion processes. The maximal amount of waste lipids that can be sustainably accommodated, and effectively converted to methane in anaerobic reactors, is limited by several problems including adsorption, sludge flotation, washout, and inhibition. These difficulties can be circumvented by appropriate feeding, mixing, and solids separation strategies, provided by suitable reactor technology and operation. In recent years, membrane bioreactors and flotation-based bioreactors have been developed to treat lipid-rich wastewater. In parallel, the increasing knowledge on the diversity of complex microbial communities in anaerobic sludge, and on interspecies microbial interactions, contributed to extend the knowledge and to understand more precisely the limits and constraints influencing the anaerobic biodegradation of lipids in anaerobic reactors. This critical review discusses the most important principles underpinning the degradation process and recent key discoveries and outlines the current knowledge coupling fundamental and applied aspects. A critical assessment of knowledge gaps in the field is also presented by integrating sectorial perspectives of academic researchers and of prominent developers of anaerobic technology. © 2022 American Chemical Society. All rights reserved.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Preliminary Assessment of Pyrolysis Biochar Derived from Milk/Dairy Processing Sludge as a Potential Component of Fertilizers

Kwapinska, M., Pisano, I., Leahy, J.J.

ACS Sustainable Chemistry and Engineering, 2022

Disposal of waste-activated sludge [dairy processing sludge, (DPS)] from wastewater treatment plants located in milk processing companies is an increasing concern. DPS is usually applied to farmlands in the vicinity of the dairy companies. This practice is becoming unsustainable due to uncontrolled nutrient loss and potential soil contamination. We propose to recover nutrients in the form of biochar. This paper examines the properties of biochars obtained from slow pyrolysis of DPS. DPS samples were pyrolyzed at laboratory and pilot scale at 600 and 700 °C. The elemental properties of biochars, the content of primary and secondary nutrients, as well as contaminants were examined and compared against the European Union Fertilizing Products Regulation. The biochars meet the specified limits for hydrogen-to- organic carbon ratio, chloride, and polycyclic aromatic hydrocarbons intended for gasification and pyrolysis component category materials. In six out of eight biochars, the content of phosphorus (P) as a single declared nutrient and the level of contaminants meet those required for an organo-mineral fertilizer. Only two biochars meet the required concentrations of nitrogen, phosphorus, and potassium. A minimum solid content of 30% in DPS is required to make the process of biochar production energetically sustainable.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Current Trends in Biological Valorization of Waste-Derived Biomass: The Critical Role of VFAs to Fuel A Biorefinery

Nzeteu C.; Coelho F.; Davis E.; Trego A.; O'Flaherty V.

Fermentation, 2022

The looming climate and energy crises, exacerbated by increased waste generation, are driving research and development of sustainable resource management systems. Research suggests that organic materials, such as food waste, grass, and manure, have potential for biotransformation into a range of products, including: high-value volatile fatty acids (VFAs); various carboxylic acids; bioenergy; and bioplastics. Valorizing these organic residues would additionally reduce the increasing burden on waste management systems. Here, we review the valorization potential of various sustainably sourced feedstocks, particularly food wastes and agricultural and animal residues. Such feedstocks are often micro-organism-rich and well-suited to mixed culture fermentations. Additionally, we touch on the technologies, mainly biological systems including anaerobic digestion, that are being developed for this purpose. In particular, we provide a synthesis of VFA recovery techniques, which remain a significant technological barrier. Furthermore, we highlight a range of challenges and opportunities which will continue to drive research and discovery within the field. Analysis of the literature reveals growing interest in the development of a circular bioeconomy, built upon a biorefinery framework, which utilizes biogenic VFAs for chemical, material, and energy applications. © 2022 by the authors.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Development of an enhanced chain elongation process for caproic acid production from waste-derived lactic acid and butyric acid

Nzeteu C.O.; Coelho F.; Trego A.C.; Abram F.; Ramiro-Garcia J.; Paulo L.; O'Flaherty V.

Journal of Cleaner Production, 2022

Medium chain carboxylic acids (MCCA), such as caproic acid, are high-value chemicals with many industrial applications. The development of a biological mixed culture chain elongation (CE) process, using waste-derived lactic acid offers an opportunity to sustainably produce MCCA, such as caproic acid. However, the nature of the biological conversions in mixed communities is still poorly understood. In particular, inhibition of methanogens and MCCA competitors for the development of a specialised inoculum and optimisation of lactic acid conversion and selectivity toward caproic acid are required. Here, the impacts of physical and chemical treatments of the inoculum (anaerobic granular sludge) on the efficiency of CE were evaluated. Additionally, the effects of lactic acid/butyric acid concentrations and ratio (r LA/BA) on selectivity and concentration of caproic acid were investigated. Blended and acidified (pH 3) granular sludge yielded the highest caproic acid concentrations. Moreover, using pH 3 acidified and blended granular sludge, substrate concentrations of 250 mM (r LA/BA = 1.5:1) and 300 mM (r LA/BA = 1:1) were optimal for efficient caproic acid accumulation with a selectivity ≥ 90 and a mole of lactic to mole of butyric acid consumption (C r LA/BA) of 2:1. However, a similar outcome was achieved when using an enriched caproic acid producing culture with a lower substrate concentration of 200 mM (r LA/BA = 1:1). We report a lactic acid to butyric acid threshold concentration, below and above which the selectivity toward caproic acid is reduced. With these optimum substrate concentrations and ratios, the feasibility for efficient caproic acid synthesis using lactic acid as electron donor and butyric acid as electron acceptor was established. Caproic acid synthesis was completely inhibited when the substrate concentrations were increased to 400 mM (r LA/BA = 1:1). Finally, species affiliated with Ruminococcaceae were likely involved in the caproic acid synthesis. These findings have strong potential for process-design applications in continuous reactor systems, thus allowing for continuous, efficient and sustainable caproic acid production from biomass-derived lactic acid and butyric acid. © 2022 The Authors

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Funding Source: DPTC 1

Research Programme: Pillar 5

Microbial community assembly and dynamics in Granular, Fixed-Biofilm and planktonic microbiomes valorizing Long-Chain fatty acids at 20 °C

Singh S.; Rinta-Kanto J.M.; Lens P.N.L.; Kokko M.; Rintala J.; O'Flaherty V.; Ijaz U.Z.; Collins G.

Bioresource Technology, 2022

Distinct microbial assemblages evolve in anaerobic digestion (AD) reactors to drive sequential conversions of organics to methane. The spatio-temporal development of three such assemblages (granules, biofilms, planktonic) derived from the same inoculum was studied in replicated bioreactors treating long-chain fatty acids (LCFA)-rich wastewater at 20 °C at hydraulic retention times (HRTs) of 12–72 h. We found granular, biofilm and planktonic assemblages differentiated by diversity, structure, and assembly mechanisms; demonstrating a spatial compartmentalisation of the microbiomes from the initial community reservoir. Our analysis linked abundant Methanosaeta and Syntrophaceae-affiliated taxa (Syntrophus and uncultured) to their putative, active roles in syntrophic LCFA bioconversion. LCFA loading rates (stearate, palmitate), and HRT, were significant drivers shaping microbial community dynamics and assembly. This study of the archaea and syntrophic bacteria actively valorising LCFAs at short HRTs and 20 °C will help uncover the microbiology underpinning anaerobic bioconversions of fats, oil and grease. © 2021 The Author(s)

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Funding Source: DPTC 1

Research Programme: Pillar 5

Beyond Basic Diversity Estimates—Analytical Tools for Mechanistic Interpretations of Amplicon Sequencing Data

Trego A.; Keating C.; Nzeteu C.; Graham A.; O’Flaherty V.; Ijaz U.Z.

Microorganisms, 2022

Understanding microbial ecology through amplifying short read regions, typically 16S rRNA for prokaryotic species or 18S rRNA for eukaryotic species, remains a popular, economical choice. These methods provide relative abundances of key microbial taxa, which, depending on the experimental design, can be used to infer mechanistic ecological underpinnings. In this review, we discuss recent advancements in in situ analytical tools that have the power to elucidate ecological phenomena, unveil the metabolic potential of microbial communities, identify complex multidimensional interactions between species, and compare stability and complexity under different conditions. Additionally, we highlight methods that incorporate various modalities and additional information, which in combination with abundance data, can help us understand how microbial communities respond to change in a typical ecosystem. Whilst the field of microbial informatics continues to progress substantially, our emphasis is on popular methods that are applicable to a broad range of study designs. The application of these methods can increase our mechanistic understanding of the ongoing dynamics of complex microbial communities. © 2022 by the authors.

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Research Programme: Pillar 5

Microbial electrosynthesis of acetate from CO₂ in three-chamber cells with gas diffusion biocathode under moderate saline conditions

Dessì P.; Buenaño-Vargas C.; Martínez-Sosa S.; Mills S.; Trego A.; Ijaz U.Z.; Pant D.; Puig S.; O'Flaherty V.; Farràs P.

Environmental Science and Ecotechnology, 2023

The industrial adoption of microbial electrosynthesis (MES) is hindered by high overpotentials deriving from low electrolyte conductivity and inefficient cell designs. In this study, a mixed microbial consortium originating from an anaerobic digester operated under saline conditions ($\sim 13 \text{ g L}^{-1} \text{ NaCl}$) was adapted for acetate production from bicarbonate in galvanostatic (0.25 mA cm^{-2}) H-type cells at 5, 10, 15, or $20 \text{ g L}^{-1} \text{ NaCl}$ concentration. The acetogenic communities were successfully enriched only at 5 and $10 \text{ g L}^{-1} \text{ NaCl}$, revealing an inhibitory threshold of about $6 \text{ g L}^{-1} \text{ Na}^+$. The enriched planktonic communities were then used as inoculum for 3D printed, three-chamber cells equipped with a gas diffusion biocathode. The cells were fed with CO₂ gas and operated galvanostatically (0.25 or 1.00 mA cm^{-2}). The highest production rate of $55.4 \text{ g m}^{-2} \text{ d}^{-1}$ ($0.89 \text{ g L}^{-1} \text{ d}^{-1}$), with 82.4% Coulombic efficiency, was obtained at $5 \text{ g L}^{-1} \text{ NaCl}$ concentration and 1 mA cm^{-2} applied current, achieving an average acetate production of 44.7 kg MWh^{-1} . Scanning electron microscopy and 16S rRNA sequencing analysis confirmed the formation of a cathodic biofilm dominated by *Acetobacterium* sp. Finally, three 3D printed cells were hydraulically connected in series to simulate an MES stack, achieving three-fold production rates than with the single cell at 0.25 mA cm^{-2} . This confirms that three-chamber MES cells are an efficient and scalable technology for CO₂ bio-electro recycling to acetate and that moderate saline conditions ($5 \text{ g L}^{-1} \text{ NaCl}$) can help reduce their power demand while preserving the activity of acetogens. © 2023 The Authors

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Funding Source: DPTC 1

Research Programme: Pillar 5

Microbial community response to temperature reduction during anaerobic treatment of long chain fatty acids-containing wastewater

Liu Y.-C.; Ramiro-Garcia J.; O'Connor S.; Paulo L.M.; Maria Braguglia C.; Cristina Gagliano M.; O'Flaherty V.

Bioresource Technology, 2024

Acclimating mesophilic biomass to low temperatures have been used to start-up psychrophilic anaerobic reactors, but limited microbial information is available during the acclimation. To investigate microbial responses to temperature reductions, duplicate lab-scale anaerobic digestion (AD) reactors were operated for 166 days, with the temperature being reduced from 37°C to 15°C, using synthetic long chain fatty acid (LCFA)-containing wastewater as the feedstock. The acclimated biomass at 15°C exhibited efficient removal of organic matter (total COD>75%, soluble COD>88%, and LCFA>99%). Temperature reductions lead to significant reductions in microbiome diversity. Fermentative bacteria were highly dynamic and functional redundant during temperature reductions. Smithella was the dominant syntrophic bacteria involved in LCFA degradation coupled with Methanotherix and Methanocorpusculum at 15°C. Membrane modifications and compatible cellular solutes production were triggered by temperature reductions as microbial response to cold stress. This study provided molecular insights in microbial acclimation to low temperatures for psychrophilic AD. © 2024 The Author(s)

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Funding Source: DPTC 1

Research Programme: Pillar 5

Unifying concepts in methanogenic, aerobic, and anammox sludge granulation

Mills S.; Trego A.C.; Prevedello M.; De Vrieze J.; O'Flaherty V.; Lens P.N.L.; Collins G.

Environmental Science and Ecotechnology, 2024

The retention of dense and well-functioning microbial biomass is crucial for effective pollutant removal in several biological wastewater treatment technologies. High solids retention is often achieved through aggregation of microbial communities into dense, spherical aggregates known as granules, which were initially discovered in the 1980s. These granules have since been widely applied in upflow anaerobic digesters for waste-to-energy conversions. Furthermore, granular biomass has been applied in aerobic wastewater treatment and anaerobic ammonium oxidation (anammox) technologies. The mechanisms underpinning the formation of methanogenic, aerobic, and anammox granules are the subject of ongoing research. Although each granule type has been extensively studied in isolation, there has been a lack of comparative studies among these granulation processes. It is likely that there are some unifying concepts that are shared by all three sludge types. Identifying these unifying concepts could allow a unified theory of granulation to be formed. Here, we review the granulation mechanisms of methanogenic, aerobic, and anammox granular sludge, highlighting several common concepts, such as the role of extracellular polymeric substances, cations, and operational parameters like upflow velocity and shear force. We have then identified some unique features of each granule type, such as different internal structures, microbial compositions, and quorum sensing systems. Finally, we propose that future research should prioritize aspects of microbial ecology, such as community assembly or interspecies interactions in individual granules during their formation and growth. © 2023 The Authors

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Funding Source: DPTC 1

Research Programme: Pillar 5

Full-scale study on high-rate low-temperature anaerobic digestion of agro-food wastewater: process performances and microbial community

Paulo L.M.; Liu Y.-C.; Castilla-Archilla J.; Ramiro-Garcia J.; Hughes D.; Mahony T.; Conall Holohan B.; Wilmes P.; O'Flaherty V.

Water Science and Technology, 2024

The fast-growing global population has led to a substantial increase in food production, which generates large volumes of wastewater during the process. Despite most industrial wastewater being discharged at lower ambient temperatures (<20 °C), majority of the high-rate anaerobic reactors are operated at mesophilic temperatures (>30 °C). High-rate low-temperature anaerobic digestion (LtAD) has proven successful in treating industrial wastewater both at laboratory and pilot scales, boasting efficient organic removal and biogas production. In this study, we demonstrated the feasibility of two full-scale high-rate LtAD bioreactors treating meat processing and dairy wastewater, and the microbial communities in both reactors were examined. Both reactors exhibited rapid start-up, achieving considerable chemical oxygen demand (COD) removal efficiencies (total COD removal >80%) and generating high-quality biogas (CH₄ in biogas >75%). Long-term operations (6-12 months) underscored the robustness of LtAD bioreactors even during winter periods (average temperature <12 °C), as evidenced by sustained high COD removal rates (total COD removal >80%). The stable performance was underpinned by a resilient microbial community comprising active acetoclastic methanogens, hydrolytic, and fermentative bacteria. These findings underscore the feasibility of high-rate low-temperature anaerobic wastewater treatment, offering promising solutions to the zero-emission wastewater treatment challenge. © 2024 The Authors.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Individual methanogenic granules are whole-ecosystem replicates with reproducible responses to environmental cues

Trego A.; O'Sullivan S.; O'Flaherty V.; Collins G.; Ijaz U.Z.

Environmental Microbiome, 2024

Background: In this study, individual methanogenic (anaerobic), granular biofilms were used as true community replicates to assess whole-microbial-community responses to environmental cues. The aggregates were sourced from a lab-scale, engineered, biological wastewater treatment system, were size-separated, and the largest granules were individually subjected to controlled environmental cues in micro-batch reactors (μ BRs). **Results:** Individual granules were identical with respect to the structure of the active community based on cDNA analysis. Additionally, it was observed that the active microbial community of individual granules, at the depth of 16S rRNA gene sequencing, produced reproducible responses to environmental changes in pH, temperature, substrate, and trace-metal supplementation. We identified resilient and susceptible taxa associated with each environmental condition tested, as well as selected specialists, whose niche preferences span the entire trophic chain required for the complete anaerobic degradation of organic matter. **Conclusions:** We found that single anaerobic granules can be considered highly-replicated whole-ecosystems with potential usefulness for the field of microbial ecology. Additionally, they act as the smallest whole-community unit within the meta-community of an engineered bioreactor. When subjected to various environmental cues, anaerobic granules responded reproducibly allowing for rare or unique opportunities for high-throughput studies testing whole-community responses to a wide range of environmental conditions. © The Author(s) 2024.

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Funding Source: DPTC 1

Research Programme: Pillar 5

Microbiomes of high-rate anaerobic digestors reveal ‘Study’-specific factors and limitations of synthetic wastewater

Keating C.; Trego A.; O'Flaherty V.; Ijaz U.Z.

Water Research, 2025

Anaerobic digestion (AD) is a key technology for the treatment of organic wastes and the production of renewable energy. The stability of the process hinges on the underlying microbial populations. Amplicon sequencing is increasingly used to characterise AD microbiomes, yet sequencing efforts have not translated to process engineering of the microbiome or prediction of failure using microbial tools. Using high-rate biofilm wastewater bioreactors as a study system, we aimed to i) discern trends in archaeal and bacterial diversity, ii) identify a core AD microbiome, iii) determine the functional stability of AD microbiomes, and iv) correlate taxa to experimental conditions. We analysed amplicon sequencing data from 32 high-rate anaerobic digester studies (> 1258 samples) at various operational conditions and applied a suite of statistical microbiome tools. We found that taxonomic archaeal diversity was highly study dependent, while functional diversity was highly shared across studies. A core AD microbiome was identified with > 100 bacterial genera and 6 archaeal genera which were present at > 1 % relative abundance in at least 50 % of samples. Interestingly, we observed that microbiome stability was significantly impacted by the choice of real or synthetic wastewater, with synthetic wastewaters yielding a more stable and less complex microbiome. This was correlated to the abundances of 37 taxa in the synthetic wastewater, including 3 key methanogens (Methanotherix, Methanobacterium, and Methanosphaerula). This suggests that when synthetic wastewater is used in experimental studies, it may not result in an AD microbiome representative of real wastewater treatment systems. © 2025

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Competitively-Won Funding Publications: CareerFIT

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Transformation of organic contamination from wastewater into bioplastics (polyhydroxyalkanoate) by microorganisms

Fra-Vázquez A.; Palmeiro-Sánchez T.; Del Río A.; Mosquera-Corral A.

Wastewater Treatment Residues as Resources for Biorefinery Products and Biofuels, 2019

Biopolymers like polyhydroxyalkanoates (PHA) are potential candidates to substitute conventional plastics produced from petroleum. PHA are polymers with plastic properties and produced from mixed microbial cultures and renewable sources, like wastewater. Thus, they are biobased and biodegradable and named as bioplastics. In general, wastewater containing biodegradable organic matter is suitable to be used for PHA production. Wastewater from different industries (mainly from the agrofood sector) has been evaluated as substrate to produce PHA. Results indicate that between 6% and 65% of the organic matter contained in the wastewater is transformed into PHA. Different bacterial populations present in the activated sludge have the ability to accumulate these PHA in transient conditions, for example, of the presence-absence of organic matter or the so-called aerobic dynamic feeding (ADF). Although the use of wastewater and microbial mixed cultures to produce PHA is increasing in interest due to the substantial reduction of costs associated to the use of a waste and the required nonsterile conditions, several challenges have to be faced before the process scale up. The increase of volume productivity and the reduction of costs during the PHA extraction step are among them. © 2020 Elsevier Inc. All rights reserved.

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Research Programme: CareerFIT

Heat and chemical treatments affect the viability, morphology, and physiology of *Staphylococcus aureus* and its subsequent antibody labeling for flow cytometric analysis

Kennedy, Deirdre, Cronin, Ultan P., Piterina, Anna, Wilkinson, Martin G.

Applied and Environmental Microbiology, 2019

The effects of heat and chemical treatments on *Staphylococcus aureus* viability and physiology and their subsequent effects on antibody binding ability and cell morphology were measured. Treatments included lethal and sublethal heat; exposure to organic acids, salt, and sodium hydroxide; and freeze-thawing. Strain-related differences in viability were noted depending on treatment and were reflected in changes in physiology as monitored by flow cytometry (FCM) using three different staining protocols: SYTO 9/propidium iodide (PI), DiOC2(3), or calcein acetoxymethyl ester (calcein-AM)/PI. Treatments that resulted in significant losses in viability as measured by plate counting were reflected better by the first two staining combinations, as intracellular calcein-AM uptake may have been impaired by certain treatments. FCM analysis using labeling by commercial anti-*S. aureus* antibodies indicated that differences in cell physiology as a result of treatments influenced immunofluorescence detection. The ratio of the mean fluorescence intensities of stained cells to those of unstained cells [MFI/MFI(us)] varied with treatment, five of these treatments, including freeze-thaw, citric acid, oxalic acid, NaCl, and NaOH treatments, resulted in significantly lower fluorescence values compared to controls.

IMPORTANCE FCM data indicated that cells conventionally considered to be dead and which would not give rise to CFU in a plate count assay, e.g., cells heated to 80°C, were labeled by antibody staining. This finding suggests that without the inclusion of a live/dead discriminating dye, these cells would be erroneously detected as viable within an FCM assay. Reductions in antibody staining due to physicochemical treatment were strain related, reflecting the complexity of the phenomenon under study and illustrating that substantial validation of any new antibody detection-based method, including physiological staining and cell sorting, should be undertaken. Researchers should be aware of physicochemical treatments causing false-negative results: in this study, freeze-thawing severely reduced antibody binding without affecting the viability of a substantial percentage of cells. Scanning electron microscopy carried out on treated cells revealed a range of morphological changes resulting from physicochemical treatments which may have hindered antibody binding.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Byproducts from Dairy Processing

Oliveira, D., Fox, P., O'Mahony, J.A.

Byproducts from Agriculture and Fisheries: Adding Value for Food, Feed, Pharma and Fuels,
2019

In the processing of milk, what might be considered a byproduct is arbitrary in some cases, e.g. in the manufacture of butter or butter oil, buttermilk and butter serum are clearly byproducts, whereas, in the separation of milk, either the cream or the skimmed milk could be the byproduct, depending on the objective of the process. This chapter considers cream and skimmed milk as byproducts. In dairy processing, the term permeate is used to describe the fraction of milk or its derived streams which can permeate through the selectively-permeable membranes used for fractionation, enrichment or purification of target nutrients using pressure-driven membrane filtration processing. Whey permeate, as a byproduct of whey processing, has been a dairy processing side stream since the introduction of UF technology in the 1960s for the removal and concentration of proteins from whey. The salts of milk are generally recovered as a component of dairy products.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Delactosed permeate as a dairy processing co-product with major potential value: a review

Oliveira, D., Puri, R., Fenelon, M.A., O'Mahony, J.A.

International Journal of Food Science and Technology, 2019

Delactosed permeate (DLP) is the co-product generated during the separation of pre-crystallised lactose from milk and whey permeates. DLP production has grown with the increased production of high protein content ingredients such as whey protein concentrates and isolates. Although DLP is nutritionally rich, with approximately 0.5–1.5, 68–70, 9–10 and 8–9 g/100 g dry matter of protein, total sugars, total mineral and organic acids, respectively, it is still currently underutilised, mostly for animal feed or energy production. There are a number of novel, promising and sustainable DLP-derived food and non-food applications which are the subject of current research. Therefore, there exists the opportunity to exploit this dairy co-product in the development of new value-added ingredients. In this comprehensive review, DLP production, processing challenges and potential applications are discussed, along with identification and assessment of selected strategies for the valorisation of DLP.

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Research Programme: CareerFIT

High-Yield Synthesis of Poly(3-hydroxybutyrate- co-3-hydroxyvalerate) Copolymers in a Mixed Microbial Culture: Effect of Substrate Switching and F/M Ratio

Palmeiro-Sánchez T.; Val Del Rio A.; Fra-Vázquez A.; Luis Campos J.; Mosquera-Corral A.

Industrial and Engineering Chemistry Research, 2019

The accumulation capacity of a mixed microbial culture (MMC) is affected if the substrate used in the accumulation experiments differs from the one used in the enrichment. For this reason, the effect of substrate switching was studied to determine the versatility of an MMC enriched in a mixture of volatile fatty acids (VFAmix) to overcome this problem. The MMC was enriched using a VFAmix composed of 48.3:24.3:7.3:14.7 Cmm acetic (HAc), propionic (HPr), butyric (HBu), and valeric (HVa) acids, respectively. The accumulation capacity was tested using single VFAs (HAc, HPr, HBu, and HVa), as well as the VFAmix used in the enrichment. The accumulation capacities were 52.8 ± 4.7 , 48.8 ± 4.3 , 45.2 ± 3.0 , 48.4 ± 1.0 , and 54.5 ± 8.0 wt% for HAc, HPr, HBu, HVa, and VFAmix, respectively, with polymer compositions of 50.6 ± 15.1 , 0.4 ± 0.1 , 63.2 ± 1.5 , 0.3 ± 0.0 , and 2.0 ± 0.7 g 3-HB/g 3-HV, following the same order. The average yields were 0.84 ± 0.08 , 0.76 ± 0.09 , 0.74 ± 0.02 , 0.70 ± 0.01 , and 0.68 ± 0.09 CmolPHA/CmolVFA for HVa, VFAmix, HAc, HBu, and HPr, respectively. The feed-to-microorganism (F/M) ratio showed that values of 1-7 CmolVFA/(CmolX) in the accumulation experiments led to the optimal yields. Based on the results obtained, it seems feasible to enrich an MMC able to produce tailor-made biopolymers from different VFAs at high yields. © 2019 American Chemical Society.

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Research Programme: CareerFIT

Protein a-mediated binding of staphylococcus spp. To antibodies in flow cytometric assays and reduction of this binding by using Fc receptor blocking reagent

Cronin U.P.; Girardeaux L.; O'Meara E.; Wilkinson M.G.

Applied and Environmental Microbiology, 2020

Staphylococcus aureus and other coagulase-positive Staphylococcus spp. bind the Fc region of IgG antibodies through expression of protein A (SpA). These species have consequently been a source of false-positive signals in antibody-based assays designed to detect other target bacteria. Here, flow cytometry was used to study the influence of a number of factors on the SpA-mediated binding of single cells to an anti-human IgG antibody, including strain, heat killing, overnight storage, growth phase, cell physiology, surface adhesion, and growth in model food systems. Through the costaining of antibody-stained cells with the permeability dye propidium iodide and calcein violet AM, the cell physiological status was related to SpA-mediated antibody binding. Generally, permeabilized cells lacking esterase activity did not strongly bind antibody. The binding of a number of commercially available polyclonal IgG antibodies to non-Staphylococcus spp. was also characterized. Not all SpA-expressing species showed strong binding of mouse IgG, and one species not known to express SpA showed strong binding. Most SpA-expressing strains bound rabbit IgG antibodies to some extent, whereas only one strain bound goat IgG. To reduce or eliminate SpA-mediated IgG binding, the following products were evaluated as blocking reagents and applied prior to staining with primary or secondary antibody: normal rabbit serum, mouse IgG isotype control, goat IgG, and a commercial FcR blocking reagent. Only the FcR blocking reagent consistently reduced SpA-mediated binding of Staphylococcus spp. to antibodies against other species and could be recommended as a blocking reagent in immunoassays designed to detect non-Staphylococcus species. **IMPORTANCE** This study characterizes a widespread but little-studied problem associated with the antibody-based detection of microbes-the Staphylococcus protein A (SpA)-mediated binding of IgG antibodies-and offers a solution: the use of commercial FcR blocking reagent. A common source of false-positive signals in the detection of microbes in clinical, food, or environmental samples can be eliminated by applying this study's findings. Using flow cytometry, the authors demonstrate the extent of heterogeneity in a culture's SpA-mediated binding of antibodies and that the degree of SpA-mediated antibody binding is strain, growth phase, and food matrix dependent and influenced by simulated food processing treatments and cell adherence. In addition, our studies of SpA-mediated binding of Staphylococcus spp. to antibodies against other bacterial species produced a very nuanced picture, leading us to recommend testing against multiple strains of *S. aureus* and *S. hyicus* of all antibodies to be incorporated into any immunoassay designed to detect a non-Staphylococcus spp. © 2020 Cronin et al.

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Research Programme: CareerFIT

Gaps in the assortment of rapid assays for microorganisms of interest to the dairy industry

O'Grady J.; Cronin U.; Tierney J.; Piterina A.V.; O'Meara E.; Wilkinson M.G.

Advances in Applied Microbiology, 2020

This review presents the results of a study into the offering of rapid microbial detection assays to the Irish dairy industry. At the outset, a consultation process was undertaken whereby key stakeholders were asked to compile a list of the key microorganisms of interest to the sector. The resultant list comprises 19 organisms/groups of organisms divided into five categories: single pathogenic species (*Cronobacter sakazakii*, *Escherichia coli* and *Listeria monocytogenes*); genera containing pathogenic species (*Bacillus*, *Clostridium*, *Listeria*, *Salmonella*; *Staphylococcus*); broad taxonomic groupings (Coliforms, Enterobacteriaceae, fecal Streptococci, sulfite reducing bacteria/sulfite reducing Clostridia [SRBs/SRCs], yeasts and molds); organisms displaying certain growth preferences or resistance as regards temperature (endospores, psychrotrophs, thermodurics, thermophiles); indicators of quality (total plate count, *Pseudomonas* spp.). A survey of the rapid assays commercially available for the 19 organisms/groups of organisms was conducted. A wide disparity between the number of rapid tests available was found. Four categories were used to summarize the availability of rapid assays per organism/group of organisms: high coverage (> 15 assays available); medium coverage (5–15 assays available); low coverage (< 5 assays available); no coverage (0 assays available). Generally, species or genera containing pathogens, whose presence is regulated-for, tend to have a good selection of commercially available rapid assays for their detection, whereas groups composed of heterogenous or even undefined genera of mainly spoilage organisms tend to be “low coverage” or “no coverage.” Organisms/groups of organisms with “low coverage” by rapid assays include: *Clostridium* spp.; fecal Streptococci; and *Pseudomonas* spp. Those with “no coverage” by rapid assays include: endospores; psychrotrophs; SRB/SRCs; thermodurics; and thermophiles. An important question is: why have manufacturers of rapid microbiological assays failed to respond to the necessity for rapid methods for these organisms/groups of organisms? The review offers explanations, ranging from the technical difficulty involved in detecting as broad a group as the thermodurics, which covers the spores of multiple sporeforming genera as well as at least six genera of mesophilic nonsporeformers, to the taxonomically controversial issue as to what constitutes a fecal Streptococcus or SRBs/SRCs. We review two problematic areas for assay developers: validation/certification and the nature of dairy food matrices. Development and implementation of rapid alternative test methods for the dairy industry is influenced by regulations relating to both the microbiological quality standards and the criteria alternative methods must meet to qualify as acceptable test methods. However, the gap between the certification of developer's test systems as valid alternative methods in only a handful of representative matrices, and the requirement of dairy industries to verify the performance of alternative test systems in an extensive and diverse range of dairy matrices needs to be bridged before alternative methods can be widely accepted and adopted in the dairy industry. This study concludes that many important dairy matrices have effectively been ignored by assay developers. © 2020 Elsevier Inc.

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Research Programme: CareerFIT

Composition, Fractionation, Techno-Functional Properties and Applications of Milk Fat Globule Membrane–Derived Material

Oliveira, D., O'Mahony, J.A.

Advanced Dairy Chemistry: Vol.2 Lipids, 2020

Milk fat occurs as globules with a non-polar lipid core composed primarily of triglycerides and surrounded by the milk fat globule membrane (MFGM). Because of its cellular membrane origin, the MFGM is the richest source of phospholipids (PLs), glycolipids, gangliosides and glycoproteins in milk. Skimmed milk, buttermilk and butter serum are especially enriched with respect to these components. Due to their origin, structure and original function in stabilising the fat globules in whole milk, MFGM-derived materials are efficient and natural emulsifiers in addition to their health benefits. However, emulsifying properties of these MFGM-derived materials are strongly dependent on their content and profile of polar lipids and proteins, and the utilisation of MFGM to formulate PL-enriched ingredients remains poorly exploited commercially. This chapter aims to compile the available information on MFGM composition, particularly PLs and specific membrane proteins. The influence of the processing steps, the techno-functional properties and potential applications of MFGM-derived material are also discussed.

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Research Programme: CareerFIT

Novel application of confocal Raman microscopy to determine the microstructure of fermented dairy products including the spatial distribution of proteins, lipids and carbohydrates

Pax, A.P.; Sheehan, J.J.

Biomedical Spectroscopy and Imaging, 2020

Background: The distribution of components in fermented dairy products forms the microstructure which influences final product texture and taste. Confocal Raman microscopy may provide new molecular information on product structure not possible with other advanced microscopy techniques.

Objective: Dairy products including non-fat and full fat yoghurt, Camembert and Cheddar cheese samples were surveyed and the product microstructure observed using confocal Raman microscopy in order to determine the applicability of the technique to dairy product analysis.

Methods: Confocal Raman microscopy provided spatially resolved chemical information on the components of fermented dairy products. In conjunction with component analysis and exploratory data analysis, spatially resolved chemical information on the components of fermented dairy products was obtained and compared.

Results: Yoghurts with differing fat levels displayed different microstructures, consistent with other techniques. The influence of different molecular structures on the Camembert cheese centre and surface was revealed and Raman microscopy also gave new insights on the chemical structures within Cheddar cheese.

Conclusions: The method provides a new technique for observing the contribution of different components to the product microstructure that may be used to monitor product quality and guide product development.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Influence of Processing Temperature on Membrane Performance and Characteristics of Process Streams Generated during Ultrafiltration of Skim Milk

Puri, R., Singh, U., O'Mahony, J.A.

Foods, 2020

The effects of processing temperature on filtration performance and characteristics of retentates and permeates produced during ultrafiltration (UF) of skim milk at 5, 20, and 50 °C were investigated. The results indicate that despite higher flux at 50 °C, UF under these conditions resulted in greater fouling and rapid flux decline in comparison with 5 and 20 °C. The average casein micelle diameter was higher in retentate produced at 5 and 20 °C. The retentate analysed at 5 °C displayed higher viscosity and shear thinning behaviour as compared to retentate analysed at 20 and 50 °C. Greater permeation of calcium and phosphorus was observed at 5 and 20 °C in comparison with 50 °C, which was attributed to the inverse relationship between temperature and solubility of colloidal calcium phosphate. Permeation of α -lactalbumin was observed at all processing temperatures, with permeation of β -lactoglobulin also evident during UF at 50 °C. All UF retentates were shown to have plasmin activity, while lower activity was measured in retentate produced at 5 °C. The findings revealed that UF processing temperature influences the physicochemical, rheological, and biochemical properties of, and thereby govern the resulting quality and functionality of, retentate- and permeate-based dairy ingredients.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Influence of chaperone-like activity of caseinomacropeptide on the gelation behaviour of whey proteins at pH 6.4 and 7.2.

Gaspard, S., Sharma, P., Fitzgerald, C., Tobin, J., O'Mahony, J.A., Kelly, A., Brodkorb, A.

FOOD HYDROCOLLOIDS, 2021

The effect of caseinomacropeptide (CMP) on the heat-induced denaturation and gelation of whey proteins (2.5–10%, w/v) at pH 6.4 and 7.2, at a whey protein: CMP ratio of 1:0.9 (w/w), was investigated using differential scanning calorimetry (DSC), oscillatory rheology (90 °C for 20 min) and confocal microscopy. Greater frequency-dependence in the presence of CMP suggested that the repulsive interactions between CMP and the whey proteins affected the network generated by the non-heated whey protein samples. At pH 6.4 or 7.2, CMP increased the temperature of denaturation of β -lactoglobulin by up to 3 °C and increased the gelation temperature by up to 7 °C. The inclusion of CMP strongly affected the structure of the heat-induced whey protein gels, resulting in a finer stranded structure at pH 6.4 and 7.2. The presence of CMP combined with a lower heating rate (2 °C/min) prevented the formation of a solid gel of whey proteins after heating for 20 min at 90 °C and at pH 7.2. These results show the potential of CMP for control of whey protein denaturation and gelation.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Investigation of the flowability, thermal stability and emulsification properties of two milk protein concentrates having different levels of native whey proteins

Khalesi M.; FitzGerald R.J.

Food Research International, 2021

Milk protein concentrate-85 (MPC85) is a dairy ingredient which has a diverse range of applications in food products. The technofunctional properties of two MPC85 samples having similar gross composition but different levels of native whey protein (WP), i.e., MPC85S1 and MPC85S2 with 16.6 and 6.0 g native WP/100 g protein, respectively, were compared. Rheometric analysis showed that under an applied normal stress of 1.0–15.0 kPa, the compressibility, the air permeability and the cohesiveness of MPC85S2 was higher compared to MPC85S1. Differential scanning calorimetry showed that protein denaturation in MPC85S1 began at 63 °C while for MPC85S2 it began at 70 °C. The heat coagulation time (HCT at 140 °C) for 4.2% (w/v, on a protein basis) reconstituted MPC85S1 and MPC85S2 was 2.2 and 2.7 min, respectively. While a higher lightness for MPC85S1 was evidenced using colourimeter analysis, the colour stability on oven drying at 95 °C for MPC85S2 was higher than MPC85S1. The emulsion produced with MPC85S1 flocculated after 1 d and phase separation occurred after 14 d. In the case of MPC85S2, flocculation began after 4 d while phase separation was observed at 33 d. The viscosity of MPC85S2 (4.2% (w/v) protein) was higher than MPC85S1. This study showed differences between the flowability, viscosity, colour properties, thermal stability (in powder and in reconstituted format), emulsification and buffering capacity for MPC samples having two different levels of WP denaturation. The results demonstrated that the MPCs studied having two different levels of WP denaturation could be targeted for different functional applications. The minimal/maximum level of denaturation required to induce technofunctional property differences requires further study. © 2021 The Author(s)

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Research Programme: CareerFIT

Physicochemical properties and water interactions of milk protein concentrate with two different levels of undenatured whey protein

Khalesi M.; FitzGerald R.J.

Colloids and Surfaces A: Physicochemical and Engineering Aspects, 2021

Two bovine milk protein concentrate (MPC85) samples containing 85% protein were produced using different heat treatments. The physicochemical and water absorption properties (i.e., the solubility, wettability, water holding capacity (WHC) and particle size on reconstitution) of these two MPC85 samples with two different levels of undenatured whey protein (WP, i.e., MPC85S1 and MPC85S2) were investigated. The undenatured WP level in MPC85S1 and MPC85S2 was 16.6 and 6.0 g/100 g total protein, respectively. The calcium ion activity of MPC85S1 and MPC85S2 was 2.93 and 2.64 mmol/L, respectively ($p < 0.05$). The surface elemental profile, as determined using X-ray photoelectron spectroscopy, showed that protein (84%) and lipid (16%) were the dominant macromolecules on the surface of both MPC85 powder particles. Scanning electron microscopy analysis showed that MPC85S1 powder particles were relatively spherical while MPC85S2 particles appeared agglomerated. Particle size analysis of 5% aqueous suspensions showed a broader size distribution for MPC85S1 compared to MPC85S2. The cold solubility of the samples rehydrated at 4 °C after 192 h storage at 4 °C was higher for MPC85S1 than MPC85S2, while on increasing the temperature of rehydration from 4° to 75 °C the solubility increased for both MPC85 samples. A lower contact angle, and thus higher wettability was observed for MPC85S2 in comparison to MPC85S1. While the MPC85 samples studied herein had similar gross composition, major differences in the level of heat induced WP denaturation (~2.5 times) and a 10% difference in ionic calcium content existed. Consequently differences in the ionic calcium and more importantly the differences in the interactions between casein and native/denatured WP are considered to have resulted in altered physicochemical properties and thereby different aqueous phase interactions. © 2021 The Authors

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Research Programme: CareerFIT

Bioconversion of organic pollutants in fish-canning wastewater into volatile fatty acids and polyhydroxyalkanoate

Palmeiro-Sánchez T.; Campos J.L.; Mosquera-Corral A.

International Journal of Environmental Research and Public Health, 2021

The wastewater from the cookers of a tuna-canning plant was used as feedstock for the process. It was acidified in a continuous stirred tank reactor (CSTR) of 1.5 L to produce a mixture of volatile fatty acids (VFAs). The effluent contained 28.3 ± 8.7 g CODS/L and 25.0 ± 4.6 g CODVFA/L, 4.4 ± 1.6 g NH₄⁺/L, and 10.9 ± 4.0 g Na⁺/L, which corresponds to about 28 g NaCl/L approximately. This was used to feed a PHA production system. The enriched MMC presented a capacity to accumulate PHAs from the fermented tuna wastewater. The maximum PHA content of the biomass in the fed-batch (8.35 wt% PHA) seemed very low, possibly due to the variable salinity (from 2.2 up to 12.3 g NaCl/L) and the presence of ammonium (which promoted the biomass growth). The batch assay showed a PHA accumulation of 5.70 wt% PHA, but this is a much better result if the productivity of the reactor is taken into account. The fed-batch reactor had a productivity of 10.3 mg PHA/(L h), while the batch value was about five times higher (55.4 mg PHA/(L h)). At the sight of the results, it can be seen that the acidification of fish-canning wastewater is possible even at high saline concentrations (27.7 g NaCl/L). On the other hand, the enrichment and accumulation results show us promising news and which direction has to be followed: PHAs can be obtained from challenging substrates, and the feeding mode during the accumulation stage has an important role to play when it comes to inhibition. © 2021 by the authors. Licensee MDPI, Basel, Switzerland.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Influence of transglutaminase crosslinking on casein protein fractionation during low temperature microfiltration

Puri, R., Bot, F., Singh, U., O'Mahony, J.A.

Foods, 2021

Low temperature microfiltration (MF) is applied in dairy processing to achieve higher protein and microbiological quality ingredients and to support ingredient innovation; however, low temperature reduces hydrophobic interactions between casein proteins and increases the solubility of colloidal calcium phosphate, promoting reversible dissociation of micellar β -casein into the serum phase, and thus into permeate, during MF. Crosslinking of casein proteins using transglutaminase was studied as an approach to reduce the permeation of casein monomers, which typically results in reduced yield of protein in the retentate fraction. Two treatments (a) 5 °C/24 h (TA) and (b) 40 °C/90 min (TB), were applied to the feed before filtration at 5 °C, with a 0.1 μ m membrane. Flux was high for TA treatment possibly due to the stabilising effect of transglutaminase on casein micelles. It is likely that formation of isopeptide bonds within and on the surface of micelles results in the micelles being less readily available for protein-protein and protein-membrane interactions, resulting in less resistance to membrane pores and flow passage, thereby conferring higher permeate flux. The results also showed that permeation of casein monomers into the permeate was significantly reduced after both enzymatic treatments as compared to control feed due to the reduced molecular mobility of soluble casein, mainly β -casein, caused by transglutaminase crosslinking.

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Research Programme: CareerFIT

Extraction and Characterization of Protein Concentrates from Limpets (*Patella vulgata*) and Peptide Release Following Gastrointestinal Digestion

Bilir, Gurkan; Khalesi, Mohammadreza; Cermeño, Maria; Fitzgerald, Richard J.; Ekinci, Deniz

Journal of Agricultural and Food Chemistry, 2022

This study investigated the characterization of proteins from the Irish limpet (*Patella vulgata*) and assessed the in vitro biological activities of hydrolysates obtained following gastrointestinal digestion (INFOGEST) of a limpet protein concentrate (LPC). The physicochemical properties and the digestibility of the LPC were investigated, along with the angiotensin-converting enzyme (ACE) inhibition and antioxidant activities of the LPC-digested samples. All the digested samples examined outperformed the LPC in terms of activity. Peptides were identified using LC-MS/MS after digestion. A total of 38 and 19 peptides were identified in LPC-G and LPC-GI, respectively, using a database search and a de novo approach. Most of the identified peptides had hydrophobic amino acids, which may contribute to their antioxidant and ACE inhibitory activities. The findings of this study showed that LPC has high nutritional quality with good digestibility and could serve as a potential source of antioxidative and ACE inhibitory peptides following gastrointestinal digestion.

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Biofunctional, structural, and tribological attributes of GABA-enriched probiotic yoghurts containing *Lactocaseibacillus paracasei* alone or in combination with prebiotics

Garavand F.; Daly, David F.M.; Gómez-Mascaraque, Laura G.

International Dairy Journal, 2022

This study investigated some biofunctional, structural, and tribological attributes of synbiotic yoghurts produced using *Lactocaseibacillus paracasei* as probiotic, and galactofructose, inulin, soy protein isolate, and spirulina as prebiotics. The highest gamma-aminobutyric acid (GABA) production ($99.63 \mu\text{g mL}^{-1}$) and glutamic acid consumption ($98.39 \mu\text{g mL}^{-1}$) was found in spirulina-supplemented probiotic yoghurts (YSP), followed by galactofructose-supplemented probiotic yoghurts (YGF). However, YSP exhibited the lowest probiotic viability and the greatest pH drop. The biological activity of YSP, in terms of total phenolics, antioxidant potential, antihypertensive activity, and degree of hydrolysis was significantly higher than the other yoghurts. YSP showed lower friction coefficient in the high sliding velocities compared with other yoghurt samples. The best appearance and mouthfeel was rated by panellists for YSP, while the taste, texture, and overall acceptance of other yoghurts were preferred. Overall, the synbiotic yoghurts containing spirulina, and galactofructose represent a promising strategy for development of functional dairy products.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Impact of total calcium in milk protein concentrate on its interaction with the aqueous phase

Khalesi, Mohammadreza; FitzGerald, Richard J.

Colloids and Surfaces A: Physicochemical and Engineering Aspects, 2022

The impact of calcium level in milk protein concentrate (MPC80) containing 80% protein on its interaction with the aqueous phase was investigated. Partial acidification (pH 6.0) together with cation exchange processing was employed to modify the mineral content of control MPC (CMPC, having 2.87% (w/w) calcium) to yield MPCs with medium (1.57% w/w), low (1.00% w/w) and very low (0.36% w/w) calcium contents, namely MPCF1, MPCF2 and MPCF3, respectively. The total protein contents of CMPC, MPCF1, MPCF2 and MPCF3 was 85.58, 82.01, 81.27 and 79.50 (% w/w), respectively, while their undenatured whey protein (WP) contents were 6.03, 4.97, 4.62 and 3.51 g/100 g overall protein, respectively. The mineral profiles on the powder particle surfaces of the different MPCs was determined using X-ray photoelectron spectroscopy (XPS). The surface calcium and sodium contents of CMPC and MPCF3 were 0.3% and 0.0%, and 0.2% and 0.4% atomic concentration, respectively. Technofunctional property analysis showed that MPCF2 had the highest water holding capacity (3.40 g water/g protein) among the calcium reduced MPCs. The overall solubility following centrifugation of 5% (w/v, on a protein basis) aqueous reconstituted CMPC (65.82%), MPCF1 (76%), MPCF2 (83%) and MPCF3 (97%) was determined. Furthermore, nitrogen solubility for 4% (w/v, on a protein basis) aqueous suspensions at pH 4.0, 7.0 and 10.0 showed that all samples had low solubility at pH 4.0 ranging from 10% to 22%. Complete solubility was observed at pH 7.0 for MPCF2 and MPCF3 while the solubility of MPCF1 and CMPC at this pH was 77% and 85%, respectively. Complete solubility of all MPC80 samples was observed at pH 10. In conclusion, partial acidification followed by cation exchange chromatography led to the production of MPC80s with different calcium levels. These MPC80 ingredients displayed different powder particle surface compositions and had different interactions with the aqueous phase. Targeted modification of mineral/calcium content may be employed to generate MPC ingredients with different surface properties and functionalities.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Impact of variation in calcium level on the technofunctional properties of milk protein concentrate

Khalesi, Mohammadreza; FitzGerald, Richard J.

Colloids and Surfaces A: Physicochemical and Engineering Aspects, 2022

The technofunctional properties of milk protein concentrate containing 80% protein (MPC80) with different calcium contents, i.e., MPCF1, MPCF2 and MPCF3 contained 1.57%, 1.00% and 0.36% calcium, respectively, were studied. The MPC samples with reduced calcium were produced using partial acidification followed by a cation exchange process thereby replacing calcium with sodium in an MPC80 concentrate. Scanning electron microscopy analysis of MPC80 powder particles showed that the MPCF3 powder particles were more spherical than the other samples. The MPCF3 sample had the highest emulsion stability, apparent viscosity (η_{app}) and thermal stability (during heating between 110 and 140 °C). The results showed that modification of the calcium content in MPC80 using cation exchange significantly altered its microstructure, particle size distribution, apparent viscosity, thermal stability, colour properties, oil binding capacity and emulsion stability.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Insolubility in milk protein concentrates: potential causes and strategies to minimize its occurrence

Khalesi M.; FitzGerald R.J.

Critical Reviews in Food Science and Nutrition, 2022

Milk protein concentrates (MPCs), which are produced from skim milk following a series of manufacturing steps including pasteurization, membrane filtration, evaporation and spray drying, represent a relatively new category of dairy ingredients. MPC powders mainly comprise caseins and whey proteins in the same ratio of occurrence as in milk. While bovine MPCs have applications as an ingredient in several protein enriched food products, technofunctional concerns, e.g., reduced solubility and emulsification properties, especially after long-term storage, limit their widespread and consistent utilization in many food products. Changes in the surface and internal structure of MPC powder particles during manufacture and storage occur via casein-casein and casein-whey protein interactions and also via the formation of casein crosslinks in the presence of calcium ions which are associated with diminishment of MPCs functional properties. The aggregation of micellar caseins as a result of these interactions has been considered as the main cause of insolubility in MPCs. In addition, the occurrence of lactose-protein interactions as a result of the promotion of the Maillard reaction mainly during storage of MPC may lead to greater insolubility. This review focuses on the solubility of MPC with an emphasis on understanding the factors involved in its insolubility along with approaches which may be employed to overcome MPC insolubility. Several strategies have been developed based on manipulation of the manufacturing process, along with composition, physical, chemical and enzymatic modifications to overcome MPC insolubility. Despite many advances, dairy ingredient manufacturers are still investigating technical solutions to resolve the insolubility issues associated with the large-scale manufacture of MPC. © 2021 Taylor & Francis Group, LLC.

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Research Programme: CareerFIT

Chromatographic Techniques to Separate and Identify Bioactive Compounds in Microalgae

Kiani H.; Aznar R.; Poojary M.M.; Tiwari B.K.; Halim R.

Frontiers in Energy Research, 2022

Microalgae are potential sources for the sustainable production of valuable chemicals including polyphenols, pigments, and ω -3 PUFAs. However, successful exploitation of these high value compounds in the food, healthcare and pharmaceutical sectors depends greatly on their effective separation, identification, and analysis after recovery from the biomass. The findings of this review paper illustrated that chromatographic methods coupled to different types of detectors have been used as a crucial part of research on microalgal polyphenols, Omega-3 Polyunsaturated Fatty Acids (ω -3 PUFAs), and pigments production through identification, measurement, sample preparation, and purification practices. Therefore, it is important to provide a comprehensive review regarding the current research in the field. The basic operating principles, parametric optimisation and detection units of common (liquid chromatography and gas chromatography) and novel chromatographic techniques (counter current chromatography, expanded bed adsorption chromatography and supercritical fluid chromatography) used to separate, identify, and quantify polyphenols, PUFAs and pigments from microalgae matrices are comprehensively reviewed. © 2022 Kiani, Aznar, Poojary, Tiwari and Halim.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Polyhydroxyalkanoate bio-production and its rise as biomaterial of the future

Palmeiro-Sánchez, T.; O’Flaherty, V.; Lens, P.N.L.

Journal of Biotechnology, 2022

The first observation of a polyhydroxyalkanoate (PHA) aggregate was in 1888 by Beijerinck. Despite polyhydroxybutyrate (PHB) being the first type of PHA discovered, it was not extracted and characterized until 1925 by Maurice Lemoigne in France, even before the concept of “macromolecules” was known. After more than 30 years, in 1958, Wilkinson and co-workers rediscovered PHB and its metabolic role in the cells as storage compound. PHB started to be appealing to the industry in the 1980s, when a few companies started to commercialize microbially produced PHAs. During the 1990s, the focus was on reducing production costs to make PHA production economically feasible, for instance by genetically modified microorganisms and even plants. Since then, many advances have been made: diverse wastes as feedstock, different production processes, and tailored design of biopolymers. This paper summarizes the scientific and technological development of PHAs from their discovery in 1888 until their latest applications and current commercial uses. Future perspectives have been devised too based on the current bottlenecks.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Effect of pasteurisation and foaming temperature on the physicochemical and foaming properties of nano-filtered mineral acid whey

Purwanti, Nanik; Hogan, Sean A.; Maidannyk, Valentyn A.; Mulcahy, Shane; Murphy, Eoin G.

International Dairy Journal, 2022

Foaming can pose a major challenge during processing of acid whey (AW). In this study, nano-filtered mineral AW was collected from a commercial plant before (AW0) and after pasteurisation (AWpast; 75 °C, 15 s). Both AW samples were foamed at 21 °C and in addition, AWpast was foamed at 61 °C, corresponding to the temperature of in-plant foaming. Physicochemical, foaming, and surface properties of AW samples were compared. Foaming at 21 °C resulted in less pronounced foam characteristics for AWpast compared with AW0. Pasteurisation was found not to significantly affect physicochemical properties; however, interfacial kinetics during foaming were altered, which affected foaming behaviour. Foaming of AWpast at 61 °C produced more stable, “dry” foams. FTIR spectra confirmed the influence of protein unfolding at elevated temperatures on foaming, which was reversible upon cooling. This is significant as it gives processors a mean of controlling foaming through temperature control, where possible.

Link to article: <https://doi.org/10.1016/j.idairyj.2022.105419>

Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Application of a dairy-based model system for mathematical mapping of diffusion of salt within rennet induced micellar casein concentrate matrices

Alehosseini, A., Sharma, P., Sheehan, J.

Journal of Food Engineering, 2023

Salt migration or heterogeneity within cheese blocks influences ripening and quality parameters. A finite element model, based on the unidirectional mass transfer and Fick's second law, was employed to study the influence of changing brining conditions on the diffusion coefficients of salt through rennet-induced micellar casein concentrate model systems. Increasing the calcium ion (0–0.5% w/w) and protein (7.5–15% w/w), concentrations along with decreasing salting temperature (40–10 °C) and pH levels (5.10–6.50), significantly increased salt penetration where the diffusion coefficients ranged from about 1.40×10^{-10} to 8.70×10^{-10} ($\text{m}^2 \text{s}^{-1}$). Microscopic images revealed considerable changes to the homogeneity of protein structures, wall thickness of the casein-based network channels, and levels of dead-end structures—caused by manipulating the brining conditions. Computational 3D-maps were also prepared using the simulation software to illustrate salt migration. Overall, this study showed the potential for varying specific physicochemical parameters on controlling the migration rate of salt in dairy matrices.

Link to article: <https://doi.org/10.1016/j.jfoodeng.2022.111263>

Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Non-thermal Processing of Foods: Recent Advances

Bhavya, M.L., Hebbar, H.U.

Food Engineering Series, 2023

Non-thermal processing is gaining immense popularity in food processing, as it has several advantages over thermal processing and potential to be employed in several food processing operations. The food industry is focusing on minimal or mild processing of foods which helps to address issues related to safety, with no or minimum effect on product quality to meet the growing demands of health-conscious consumers. In this regard, the advanced technologies/techniques like high pressure processing, cold plasma, light-based processing, ultrasound, membrane processing etc., have been either employed for select applications or attempted, mainly for microbial load reduction. Although, a couple of these technologies have been employed at industrial levels, there are many issues to be addressed to widen the scope of application. Detailed studies are needed in these areas to address specific issues. However, a few of the techniques are still at laboratory scale or limited to academic studies, though, the results are promising and showed potential for scale-up and commercialization. The understanding of mechanism of action of these techniques, process intervention requirements to enhance efficacy and also limitations of techniques are to be clearly taken note while expanding the scope of application. This chapter mainly focuses on critical findings of various reports, especially the recent ones, on application of novel technologies/techniques for microbial load reduction and product quality, physicochemical parameters, sensorial attributes etc.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Association between the Presence of Resistance Genes and Sanitiser Resistance of *Listeria monocytogenes* Isolates Recovered from Different Food-Processing Facilities

Cheng, Y., Mousavi, Z., Pennone, V., Hurley, D., Butler, F.

Microorganisms, 2023

Sanitisers are widely used in cleaning food-processing facilities, but their continued use may cause an increased resistance of pathogenic bacteria. Several genes have been attributed to the increased sanitiser resistance ability of *L. monocytogenes*. This study determined the presence of sanitiser resistance genes in Irish-sourced *L. monocytogenes* isolates and explored the association with phenotypic sanitiser resistance. The presence of three genes associated with sanitiser resistance and a three-gene cassette (*mdrL*, *qacH*, *emrE*, *bcrABC*) were determined in 150 *L. monocytogenes* isolates collected from Irish food-processing facilities. A total of 23 isolates contained *bcrABC*, 42 isolates contained *qacH*, one isolate contained *emrE*, and all isolates contained *mdrL*. Additionally, 47 isolates were selected and grouped according to the number and type of resistance genes, and the minimal inhibitory concentration (MIC) of these isolates for benzalkonium chloride (BAC) was determined experimentally using the broth microdilution method. The BAC resistance of the strain carrying the *bcrABC* gene cassette was significantly higher than that of strains lacking the gene cassette, and the BAC resistance of the strain carrying the *qacH* gene was significantly higher than that of strains lacking the *qacH* gene ($p < 0.05$). Isolates harbouring both the *qacH* and *bcrABC* genes did not show higher BAC resistance. With respect to environmental factors, there was no significant difference in MIC values for isolates recovered from different processing facilities. In summary, this investigation highlights the prevalence of specific sanitiser resistance genes in *L. monocytogenes* isolates from Irish food-processing settings. While certain genes correlated with increased resistance to benzalkonium chloride, the combination of multiple genes did not necessarily amplify this resistance.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

The consequence of supplementing with synbiotic systems on free amino acids, free fatty acids, organic acids, and some stability indexes of fermented milk

Garavand F.; Daly, David F.M.; Gómez-Mascaraque, Laura G.

International Dairy Journal, 2023

An investigation on the impacts of different prebiotics (inulin, galactofructose, soy protein isolate (SPI), and spirulina) and co-culturing with *Lactocaseibacillus paracasei* on the biological metabolites [free amino acids (FAAs), free fatty acids (FFAs), and organic acids] and stability parameters of fermented milk is presented. All fermented milks represented an increased FAA content compared with their milk counterparts, while the synbiotic fermented milk supplemented with galactofructose (YGF) and spirulina (YSP) were more efficient in this regard. The total organic acid content of the samples was not significantly affected by the type of prebiotics, and co-culturing by *L. paracasei*, YGF and YSP presented a different pattern, with the highest succinic acid (0.77 mm) and oxoglutaric acid (0.27 mm) contents, respectively. The thermal stability of the fermented milks did not change by loading various prebiotics and co-culturing, while the phase, colloidal, mechanical, and shear stability indexes were significantly affected.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Alginate microcapsules produced by external gelation in milk with application in dairy products

Gómez-Mascaraque L.G.; Chambon V.; Trifkovic K.; Brodkorb A.

Food Structure, 2023

This study aims to explore possibility of production of alginate microcapsules by extrusion-dripping onto bovine milk directly. As a source of calcium necessary for capsules formation, three different types of milk were used (skimmed, whole and enriched milk). In that respect, impact of milk type and properties (e.g. density, viscosity, surface tension and soluble calcium content) on capsules formation was studied. In addition, influence of alginate type and concentration on capsules formation was assessed. The properties of the milk, viscosity in particular, contributed to the greater extent of microcapsules deformation in comparison to ones produced using CaCl₂ solution. More concentrated alginate solutions (up to 1.5%), yielded in more spherical capsules; the same was noticed with G-rich alginate. Upon microcapsules production, significant decrease in soluble calcium (23–27% reduction) and total protein content (1–4% reduction) of milk was observed; this can be assigned to the interactions with alginate network, which was further confirmed via confocal laser scanning microscopy. Encapsulation efficiency study showed that developed capsules were able to entrap blue dextrans at satisfactory extent (from 43 ± 2% to 56 ± 1%), where higher efficiency of encapsulation was achieved for the blue dextrans of higher molecular weight. © 2023 Elsevier Ltd

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Improving the physical and wettability properties of skim milk powders through agglomeration and lecithination

Hailu, Y., Maidannyk, V., Murphy, E., McCarthy, N.

Journal of Food Engineering, 2023

This study aimed to reduce the bulk density of skim milk powders (SMP) and improve subsequent wettability and dissolution by a combination of agglomeration and lecithination. Agglomeration significantly increased powder particle size from a D90 of 120–201 μm , and decreased tapped bulk density (0.73–0.65 g/cm^3), although it led to increased friability (32.7%) compared to regular SMP (22.9%). Spraying lecithin on to SMP in the fluid bed improved wettability (8.94 s) compared to regular SMP (>300 s). Agglomeration without lecithination had no effect on powder wettability, similarly, adding lecithin in to liquid skim milk concentrate prior to drying did not improve subsequent powder wettability. Overall, improving the functionality of skim milk is quite complex, and while powder bulk density can be reduced by agglomeration, the particles remain susceptible to breakdown, and the wettability is relatively poor, although this can be improved by spraying lecithin directly on to the powder particles.

Link to article: <https://doi.org/10.1016/j.jfoodeng.2023.111597>

Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Contribution of whey protein denaturation to the in vitro digestibility, biological activity and peptide profile of milk protein concentrate

Khalesi, Mohammadreza; Cermeño, Maria; FitzGerald, Richard J.

Journal of Functional Foods, 2023

The impact of whey protein (WP) denaturation on the in vitro digestibility and biological activity of milk protein concentrate-85 (MPC85) was investigated. MPC85S1 and MPC85S2 having undenatured WP levels equal to 16.6 and 6.0 g/100 g overall protein, respectively, had similar in vitro protein digestibility corrected amino acid scores equal to 1.14. The samples were subjected to in vitro simulated gastrointestinal digestion while sampling was performed every 30 min during gastric (GD) followed by intestinal (GID) digestion. Liquid chromatography–mass spectroscopy showed that MPC85S1-GD, MPC85S2-GD, MPC85S1-GID and MPC85S2-GID had 50, 38, 47 and 66 unique peptides, respectively. The degree of hydrolysis, molecular mass distribution, dipeptidyl peptidase-IV inhibition, oxygen radical absorbance capacity and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging activity of the digests were compared. Overall, the results showed higher digestibility and bioactivities for low-denatured MPC85 compared to high-denatured MPC85 upon GD, however, following GID, both samples were digested to a similar extent.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

In Vitro Digestibility, Biological Activity, and Physicochemical Characterization of Proteins Extracted from Conventionally and Organically Cultivated Hempseed (*Cannabis sativa* L.)

Khalesi, Mohammadreza; Gcaza, Luthando; FitzGerald, Richard J.

Molecules, 2023

The proteins from two conventionally (CC1 and CC2) and one organically cultivated (OC) hempseed samples were extracted (by alkaline solubilization followed by isoelectric precipitation) and compared in terms of their physicochemical, digestibility and in vitro bioactivity properties. The OC hempseed had higher total protein and lower nonprotein nitrogen content. Protein extracts showed bimodal particle size distributions, with OC showing the smallest and CC1 the largest mean particle diameter ($d(0.5)$), i.e., 89.0 and 120.0 μm , respectively. Chromatographic analysis showed similar protein profiles for all three protein extracts. The protein extracts were subjected to in vitro simulated gastrointestinal digestion (SGID). Degree of hydrolysis (DH) measurement showed that the highest extent of digestion upon SGID was associated with CC1 ($11.0 \pm 1.5\%$), which also had the lowest in vitro antioxidant activity. Only the OC and OC digested samples had lipase inhibitory activity. The results indicate that the cultivation method impacted the composition, physicochemical, digestibility, and biofunctional properties of hempseed proteins.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Nitrogen and phosphate removal from dairy processing side-streams by monocultures or consortium of microalgae

Kiani, H., Azimi, Y., Li, Y., Mousavi, M., Cara, F., Mulcahy, S., McDonnell, H., Blanco, A., Halim, R.

Journal of Biotechnology, 2023

Acid-casein production generates waste streams that are rich in nitrogen (in the form of protein and nitrate) and phosphate. This makes this type of waste very difficult to treat using conventional techniques resulting in a high amount of operating cost and costly investment. In this research, the application of single culture or consortium of microalgae for uptake of nitrogen and phosphate in the wastewater of an acid-casein factory was investigated. The waste was a 1:1 mixture of nanofiltered whey permeate and dairy processing wastewater. Monocultures of *Chlorella vulgaris*, *Tetrademus obloquus*, *Nannochloropsis oenocarpa* and a consortium of the three microalgae were analyzed. The results showed that the consortium exhibited more efficient nitrogen and phosphate removal compared to the individual species. The consortium was able to rapidly hydrolyse exogenous protein present in the waste medium, removing 88% of protein and breaking down complex protein molecules into simpler compounds (such as nitrate) for assimilation into the biomass. In the first fourteen days of cultivation, the rate of nitrate assimilation by the consortium biomass was lower than that of nitrate formation from protein degradation, leading to a net increase in nitrate concentration in the medium. As protein source was depleted and biomass concentration increased, however, the rate of nitrate assimilation began to exceed that of nitrate formation allowing for net removal of nitrate. The microalgae consortium was shown to successfully bioremediate all nitrates by day 21. It was indicated that *Chlorella* and *Nannochloropsis* species were responsible for nitrogen removal in monocultures. Phosphate, on the other hand, was efficiently removed by *Tetrademus*. The results indicated that a consortium cultivation of three species of microalgae led to effective elimination of both nitrogen and phosphate. Combined flow-cytometry and microscopy analyses revealed that *Chlorella* overtook *Tetrademus* and *Nannochloropsis* to emerge as the dominant population in the consortium by the end of the cultivation cycle. It can be concluded that the application of microalgae consortium for simultaneous recovery of nitrogen and phosphate is a promising approach for treating acid-casein wastewater.

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Research Programme: CareerFIT

Enzyme-Assisted Extraction of Plant Proteins

Kleekayai, Thanyaporn, Khalesi, Mohammadreza, Amigo-Benavent, Miryam, Cermeño, Maria, Harnedy-Rothwell, Pádraigín, FitzGerald, Richard J.

Green Protein Processing Technologies from Plants: Novel Extraction and Purification Methods for Product Development, 2023

Enzyme-assisted extraction (EAE) is an environmentally friendly green processing technique used to aid protein extraction from different plant sources. This is due to its mild operating conditions, reduced waste generation and low energy consumption compared to chemical and physical extraction approaches. A range of food grade carbohydrase and protease preparations have been employed to aid protein extraction/solubilisation from different plant sources by hydrolysis of the plant cell wall and the proteins therein. Different statistical tools can be employed to optimise enzyme treatment parameters including enzyme:substrate, pH, incubation temperature and hydrolysis duration to yield maximal protein recovery. While EAE facilitates protein recovery, it may also enhance the nutritional (digestibility) and techno- and bio-functional properties of the extracted proteins, particularly when using protease-assisted extraction. Combining EAE with physical techniques, e.g., ultrasonic processing, for biomass pre-treatment can enhance plant cell wall disruption with a view to enhancing protein extraction efficiency. This approach can facilitate economic feasibility by reducing the energy required and the quantity of enzyme used and, therefore, the overall cost of the extraction process. An overview of the application of EAE in protein/peptide recovery from different plant sources including oilseeds, nuts, cereals, pulses and algae is provided herein.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Mechanism of lactose assimilation in microalgae for the bioremediation of dairy processing side-streams and co-production of valuable food products

Li, Y., Miros, S., Kiani, H., Eckhardt, H-G., Blanco, A., Mulcahy, S., McDonnell, H., Tiwari, B., Halim, R.

Journal of Applied Phycology, 2023

This study investigated the mechanism of lactose assimilation in *Nannochloropsis oceanica* 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). Mixotrophic cultivation was found to be mandatory for lactose assimilation in *N. oceanica*, with biomass production in mixotrophic cultures reaching a fourfold increase over that under heterotrophic conditions. Under mixotrophic conditions, the microalgae were able to produce β -galactosidase enzyme to hydrolyse lactose, with maximum extracellular secretion recorded on day 8 of growth cycle at 41.47 ± 0.33 U gbiomass⁻¹. No increase in the concentration of glucose or galactose was observed in the medium, confirming the ability of microalgae to indiscriminately absorb the resultant monosaccharides derived from lactose breakdown. Population analysis revealed that microalgae cells were able to maintain dominance in the mixotrophic culture, with bacteria accounting for < 12% of biomass. On the other hand, under heterotrophic conditions, native bacteria took over the culture (occupying over 95% of total biomass). The bacteria, however, were also unable to effectively assimilate lactose, resulting in limited biomass increase and negligible production of extracellular β -galactosidase. Results from the study indicate that *N. oceanica* can be effectively applied for onsite dairy wastewater treatment under strict mixotrophic conditions. This is commercially disadvantageous as it rules out the possibility of deploying heterotrophic fermentation with low-cost bioreactors and smaller areal footprint.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Cronobacter Species in the Built Food Production Environment: A Review on Persistence, Pathogenicity, Regulation and Detection Methods

Mousavi, Z.E., Hunt, K., Koolman, L., Butler, F., Fanning, S.

Microorganisms, 2023

The powdered formula market is large and growing, with sales and manufacturing increasing by 120% between 2012 and 2021. With this growing market, there must come an increasing emphasis on maintaining a high standard of hygiene to ensure a safe product. In particular, Cronobacter species pose a risk to public health through their potential to cause severe illness in susceptible infants who consume contaminated powdered infant formula (PIF). Assessment of this risk is dependent on determining prevalence in PIF-producing factories, which can be challenging to measure with the heterogeneity observed in the design of built process facilities. There is also a potential risk of bacterial growth occurring during rehydration, given the observed persistence of Cronobacter in desiccated conditions. In addition, novel detection methods are emerging to effectively track and monitor Cronobacter species across the food chain. This review will explore the different vehicles that lead to Cronobacter species' environmental persistence in the food production environment, as well as their pathogenicity, detection methods and the regulatory framework surrounding PIF manufacturing that ensures a safe product for the global consumer.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Comprehensive Genomic Characterization of *Cronobacter sakazakii* Isolates from Infant Formula Processing Facilities Using Whole-Genome Sequencing

Mousavi, Z.E., Koolman, L., Macori, G., Fanning, S., Butler, F.

Microorganisms, 2023

Cronobacter sakazakii is an opportunistic pathogen linked to outbreaks in powdered infant formula (PIF), primarily causing meningitis and necrotizing enterocolitis. Whole-genome sequencing (WGS) was used to characterize 18 *C. sakazakii* strains isolated from PIF (powdered infant formula) manufacturing plants (2011–2015). Sequence Type (ST) 1 was identified as the dominant sequence type, and all isolates carried virulence genes for chemotaxis, flagellar motion, and heat shock proteins. Multiple antibiotic resistance genes were detected, with all isolates exhibiting resistance to Cephalosporins and Tetracycline. A significant correlation existed between genotypic and phenotypic antibiotic resistance. The plasmid Col(pHAD28) was identified in the isolates recovered from the same PIF environment. All isolates harbored at least one intact phage. All the study isolates were compared with a collection of 96 publicly available *C. sakazakii* genomes to place these isolates within a global context. This comprehensive study, integrating phylogenetic, genomic, and epidemiological data, contributes to a deeper understanding of *Cronobacter* outbreaks. It provides valuable insights to enhance surveillance, prevention, and control strategies in food processing and public health contexts.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

How temperature shapes the biosynthesis of polyhydroxyalkanoates in mixed microbial cultures

Palmeiro-Sánchez, T.; Graham, A.; Lens, P.N.L.; O'Flaherty, V.

Water Environment Research, 2023

Three sequential batch reactors were operated for the enrichment in microbial communities able to store polyhydroxyalkanoates (PHAs) using activated sludge as inoculum. They ran simultaneously under the same operational conditions (organic loading rate, hydraulic and solids retention time, cycle length, C/N ratio) just with the solely difference of the working temperature: psychrophilic (15°C), mesophilic (30°C), and thermophilic (48°C). The microbial communities enriched showed different behaviors in terms of consumption and production rates. In terms of PHA accumulation, the psychrophilic community was able to accumulate an average amount of 17.7 ± 5.7 wt% poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV), the mesophilic 40.3 ± 7.0 wt% PHBV, and the thermophilic 14.8 ± 0.3 wt% PHBV in dry weight over total solids. The average PHBV production yields for each selected community were 0.41 ± 0.12 CmmolPHBV/CmmolVFA at 15°C, 0.64 ± 0.05 CmmolPHBV/CmmolVFA at 30°C, and 0.39 ± 0.14 CmmolPHBV/CmmolVFA at 48°C. The overall performance of the mesophilic reactor was better than the other two, and the copolymers obtained at this temperature contained a higher PHV fraction. The physico-chemical properties of the obtained biopolymers at each temperature were also measured, and major differences were found in the molecular weight, following an increasing trend with temperature.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Plasma-activated liquids for mitigating biofilms on food and food contact surfaces

Zhao, Y., Bhavya, M.L., Patange, A., Sun, DW, Tiwari, B.

Comprehensive Reviews in Food Science and Food Safety, 2023

Plasma-activated liquids (PALs) are emerging and promising alternatives to traditional decontamination technologies and have evolved as a new technology for applications in food, agriculture, and medicine. Contamination caused by foodborne pathogens and their biofilms has posed challenges and concerns to the food industry in terms of safety and quality. The nature of the food and the food processing environment are major factors that contribute to the growth of various microorganisms, followed by the biofilm characteristics that ensure their survival in severe environmental conditions and against traditional chemical disinfectants. PALs show an efficient impact against microorganisms and their biofilms, with various reactive species (short- and long-lived ones), physiochemical properties, and plasma processing factors playing a crucial role in mitigating biofilms. Moreover, there is potential to improve and optimize disinfection strategies using a combination of PALs with other technologies for the inactivation of biofilms. The overarching aim of this study is to build a better understanding of the parameters that govern the liquid chemistry generated in a liquid exposed to plasma and how these translate into biological effects on biofilms. This review provides a current understanding of PALs-mediated mechanisms of action on biofilms; however, the precise inactivation mechanism is still not clear and is an important part of the research. Implementation of PALs in the food industry could help overcome the disinfection hurdles and can enhance biofilm inactivation efficacy. Future perspectives in this field to expand existing state of the art to seek breakthroughs for scale-up and implementation of PALs technology in the food industry are also discussed.

Link to article: <https://doi.org/10.1111/1541-4337.13126>

Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Novel biorefinery process for extraction of laminarin, alginate and protein from brown seaweed using hydrodynamic cavitation

Zhu, X., Healy, L., Das, R., Bhavya, M.L., Karuppusamy, S., Sun, DW., O'Donnell, C.,
Tiwari, B.

Algal Research, 2023

This paper 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 maximize 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, maximizing the value of seaweed biomass. 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 up to 86.57 ± 3.72 % and 40.49 ± 2.78 % with recovery rates of 55.55 ± 3.10 % and 75.90 ± 4.49 %, respectively. Ethanol precipitation was then carried out to recover sodium alginate after the second extraction stage process using 2 % Na₂CO₃ (w/v). The sodium alginate purity extracted by employing HDC twice (HDC-HDC) was 88.98 ± 4.70 % with a recovery rate of 65.13 ± 5.14 %. The remaining residue after the biorefinery process had an enriched protein content of 17.19 ± 1.33 %. This study demonstrates that an HDC-assisted biorefinery process can significantly ($P < 0.05$) reduce energy consumption. The laminarin extracts were further characterised by antioxidant activity, anti-inflammation activity, FT-IR, and anti-microbial activity. The laminarin extracted in this study was shown to have identical bioactive activities as the commercially available samples.

Link to article: <https://doi.org/10.1016/j.algal.2023.103243>

Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Yogurt fortified with various protein hydrolysates: Texture and functional properties

Abdel-Hamid M.; Hamed A.M.; Walker G.; Romeih E.

Food Chemistry, 2024

This work evaluated the impact of incorporating 1% of commercial protein hydrolysates [rice protein hydrolysate (RPH), pea protein hydrolysate (PPH), and casein hydrolysate (CH)] on the functional, microstructure, and texture properties of set yogurt. Yogurt prepared with RPH exhibited the highest viability number of *Streptococcus thermophilus*. The addition of three hydrolysate types to yogurt revealed significant increases in the antioxidant and ACE-inhibitory activities, where the highest values were noted for the yogurt prepared with RPH. RPH exhibited no differences in texture properties (firmness, consistency, and cohesiveness) to control yogurt. These results were confirmed by scanning electron microscope examination. RPH and control yogurts showed compacted and dense structures accompanied by small pores, whereas CH and PPH yogurt structures were characterized by coarse networks with large voids. Furthermore, there was no significant impact of adding protein hydrolysates on the overall acceptability of yogurt as indicated by a sensory panel. © 2024 The Authors

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Research Programme: CareerFIT

Growth and fatty acid profile of *Nannochloropsis oceanica* cultivated on nano-filtered whey permeate

Kiani H.; Ma Q.; Xiao M.; Li Y.; Brooke F.J.; Mulcahy S.; Miros S.; Halim R.

Journal of Applied Phycology, 2024

Nano-filtered whey permeate (WP), a major by-product of dairy industry, is produced by membrane filtration of whey. The oleaginous microalga *Nannochloropsis oceanica* was successfully cultivated on WP without salinity and nutrient amendments. Growth, cell characteristics, and fatty acid profile of the cultures were analyzed using microscopy, flow cytometry, and GC analysis. WP was nitrogen limited, comprising primarily protein as a nitrogen source and only small amounts of free inorganic nitrogen (in the form of nitrate). *Nannochloropsis oceanica* (and associated bacteria) efficiently removed nitrate (100%), protein (87%), and phosphate (74%) from the whey permeate. Microscopic and flow cytometric analysis revealed diverse size distributions in whey permeate cultures, with significant cell aggregation attributed to low-salinity acclimatization and nitrogen limitation. Autofluorescence analysis revealed reduced photosynthetic activity in whey permeate-grown cells, possibly as a consequence of heightened mixotrophic activities on carbon source in the medium. Low nitrogen availability in whey permeate resulted in biomass with a fatty acid profile enriched in saturated fatty acids. Despite this, a considerable level of the omega-3 polyunsaturated fatty acid (in the form of eicosapentanoic acid or EPA) was detected at ca. 16% of total fatty acids. Whey permeate proved beneficial for the growth of *N. oceanica* and yielded high concentrations of eicosapentaenoic acid in the extracted lipids for potential applications in the feed/food industries. © The Author(s) 2024.

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Research Programme: CareerFIT

Component partitioning during microfiltration and diafiltration of whey protein concentrate in the production of whey protein isolate

Mestawet A.T.; France T.C.; Mulcahy P.G.J.; O'Mahony J.A.

International Dairy Journal, 2024

This study investigated component partitioning during cold microfiltration (MF) and diafiltration (DF) of whey protein concentrate (WPC) feed in producing whey protein isolate (WPI). Significant differences ($P < 0.05$) were found in the partitioning of components. Specifically, 60.1, 50.2, 6.32, and 75.7% of total solids, protein, fat, and ash, respectively, were partitioned into the permeate stream. Phospholipids comprised 25 and 41.4% of total fat in WPC feed and DF retentate, respectively. Sodium dodecyl sulphate-polyacrylamide gel electrophoresis showed MFGM-associated proteins were enriched in the MF retentate. The particle size in MF retentate was significantly larger ($P < 0.05$) than in the MF and DF permeate streams (450, 7.87, and 3.55 nm, respectively), providing evidence of the retention of aggregated whey proteins in the MF retentate. These findings provide an indepth understanding of component partitioning during cold MF of WPC feed, supporting the development of higher value-added ingredients. © 2024

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Microfiltration retentate co-product from whey protein isolate production - Composition, processing, applications and potential for value addition

Mestawet A.T.; France T.C.; Mulcahy P.G.J.; O'Mahony J.A.

Trends in Food Science and Technology, 2024

Background: Microfiltration retentate (MFR), also called whey protein phospholipid concentrate, is a co-product of whey protein isolate (WPI) production derived through microfiltration (MF) of whey or whey protein concentrate. Microbiological quality and protein denaturation/aggregation in the MFR stream present challenges in valorizing the stream for utilization in specialized nutritional products. As a result, MFR is underutilized, with its current applications largely limited to commodity applications in the animal feed industry as a milk replacer and in confectionery, for example. On the other hand, the production of MFR is increasing year on year due to the increase in demand for WPI with its current production representing 14–18% of the total whey processed worldwide. **Scope and approach:** In this review, we discuss MFR processing options, composition, current applications, future perspectives, and potential valorization strategies and challenges. Our approach includes a comprehensive literature review of recent studies and advancements in MFR processing. We systematically selected and analyzed peer-reviewed articles, industry websites, and reports to provide a holistic view of the current state and future directions of MFR technology. **Key findings and conclusions:** The gross chemical composition of MFR is highly variable, with typical values of fat, protein, lactose, and ash ranging from 11 to 38%, 50–70%, 1–11%, and 2–4%, respectively. The protein constituents in MFR include β -lactoglobulin, α -lactalbumin, bovine serum albumin, lactoferrin, immunoglobulins, and caseino-macropptide. Additionally, MFR is enriched with milk fat globule membrane-associated proteins such as butyrophilin, mucin 1, xanthine oxidase, and phospholipids like sphingomyelin and phosphatidylcholine. Significant research gaps exist in understanding the microbiology, bioactivity, and bioavailability of MFR components, which are crucial for supporting its valorization. Despite these gaps, there is great potential for utilizing MFR in the food industry, neonatal nutrition, and pharmaceutical applications. This potential provides opportunities to develop targeted, novel value-added ingredients from the MFR stream.

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Research Programme: CareerFIT

Alteration of Physicochemical Properties and Heating Stability of Reconstituted Acid Whey Powder by Calcium Chelating Salts

Purwanti N.; Mulcahy S.; Murphy E.G.

Food and Bioprocess Technology, 2024

Trisodium citrate (TSC) and ethylenediaminetetraacetic acid disodium salt (Na₂-EDTA) were applied in reconstituted acid whey powder (AWP) at 20% w/w, which mimicked acid whey concentration during industrial whey processing. Physicochemical properties and heat stability of the AWP suspensions with 0–50 mM TSC and Na₂-EDTA at pH 6.2 were investigated. TSC-containing suspensions prior to heating had decreasing Ca²⁺ activity, levels of sedimentation, and subtle reduction of aggregate size with increasing TSC concentrations (0–50 mM). Unheated Na₂-EDTA-containing suspensions had lower levels of sedimentation and smaller aggregate sizes than unheated TSC-containing suspensions; however, reduction of Ca²⁺ activity was only observed up to 20 mM Na₂-EDTA. Stronger effects of Na₂-EDTA than TSC on levels of sediment, viscosity, and aggregate size of AWP suspensions were observed after heating, except for 50 mM Na₂-EDTA. A remarkable difference between TSC and Na₂-EDTA addition was the nature of aggregates formed in heated suspensions. TSC-containing suspensions contained larger aggregates than corresponding Na₂-EDTA-containing suspensions, which exhibited increasing shear thinning behavior as a function of concentration. In contrast, the smaller aggregates in the corresponding Na₂-EDTA-containing suspension showed shear thickening. The inverse relationship between aggregate size and levels of sediment for TSC-containing suspensions post-heat treatment may indicate the formation of loose aggregates that resist sedimentation. © The Author(s), under exclusive licence to Springer Science+Business Media, LLC, part of Springer Nature 2023.

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Research Programme: CareerFIT

First evidence for temperature's influence on the enrichment, assembly, and activity of polyhydroxyalkanoate-synthesizing mixed microbial communities

Trego, A.; Palmeiro-Sánchez, T.; Graham, A.; Umer Zeeshan, I.; O'Flaherty, V.

Frontiers in Systems Biology, 2024

Polyhydroxyalkanoates (PHA) are popular biopolymers due to their potential use as biodegradable thermoplastics. In this study, three aerobic sequencing batch reactors were operated identically except for their temperatures, which were set at 15 °C, 35 °C, and 48 °C. The reactors were subjected to a feast–famine feeding regime, where carbon sources are supplied intermittently, to enrich PHA-accumulating microbial consortia. The biomass was sampled for 16S rRNA gene amplicon sequencing of both DNA (during the enrichment phase) and cDNA (during the enrichment and accumulation phases). All temperatures yielded highly enriched PHA-accumulating consortia. Thermophilic communities were significantly less diverse than those at low or mesophilic temperatures. In particular, *Thauera* was highly adaptable, abundant, and active at all temperatures. Low temperatures resulted in reduced PHA production rates and yields. Analysis of the microbial community revealed a collapse of community diversity during low-temperature PHA accumulation, suggesting that the substrate dosing strategy was unsuccessful at low temperatures. This points to future possibilities for optimizing low-temperature PHA accumulation. Copyright © 2024 Trego, Palmeiro-Sánchez, Graham, Ijaz and O'Flaherty.

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Research Programme: CareerFIT

Persian everlasting pea (*Lathyrus rotundifolius* L.) protein isolate as a potential protein source for food application: Effect of ultrasound-assisted extraction method on the properties of the protein isolates

Youshanlouei, Y., Kiani, H., Mousavi, M., Mousavi, Z., Tao, Y., Halim, R.

Journal of Food Process Engineering, 2024

Finding new protein sources and processes comprising targeted functional properties is crucial for the increasing global food demand. In the current paper, a new protein isolate is introduced with potential benefits based on the seeds of Persian everlasting pea (*Lathyrus rotundifolius* L.). Conventional aqueous extraction for the preparation of protein isolates (CPEP) was compared with pre-ultrasonic (PUPEP) and ultrasound-assisted (UAPEP) extractions, and the physicochemical and functional properties of the proteins were investigated. By the implication of ultrasound trials, protein recovery was increased from 49.88% for CPEP to 55.02% and 56.11%, for PUPEP and UAPEP, respectively. According to gel electrophoresis, no major distinction in molecular weight between protein fractions of different samples was perceived and sonication practice did not convert the primary structures of proteins. FTIR results uncovered changes in different extraction modes, implying that ultrasound could transform the secondary structures of the protein. Thermal properties and surface tension were decreased during sonication due to the conformational changes. The results of this study indicated that Persian everlasting pea protein isolate could be considered as a novel source of valuable protein and functional ingredient in the food industry.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Pre-treatment of whey protein concentrate using calcium sequestering salts and high-pressure homogenisation to modify component partitioning during microfiltration

Mestawet A.T.; France T.C.; Mulcahy P.G.J.; O'Mahony J.A.

International Journal of Dairy Technology, 2025

Whey protein concentrate (WPC) undergoes microfiltration (MF) to produce whey protein isolate (WPI), generating a lower value MF retentate as a co-product. Higher-than-expected protein retention in the retentate, attributed to protein aggregation, has been shown to limit WPI yield. Strategies to reverse or reduce aggregation would be expected to increase protein transmission during MF. This study investigated the effects of pre-treating WPC with 5 mM trisodium citrate (TSC), a calcium-binding salt and high-pressure homogenisation (HPH) at 650 bar, both individually and in combination, on protein transmission during MF. A WPC solution (2.4% protein) was pre-treated with TSC, HPH, TSC followed by HPH (TSC + HPH), or HPH followed by TSC (HPH + TSC). Microfiltration was performed using a 1000 kDa polyethersulfone membrane. Processing time, component partitioning and chemical composition in feed, retentate and permeate were analysed. Protein profiles were assessed using SDS-PAGE and RP-HPLC, in addition to whey protein denaturation. Data were obtained from three independent trials, with all analyses conducted in triplicate. Treatment significantly reduced processing time and increased protein permeation ($P < 0.05$). Processing time decreased by 6.4–11.0%, with TSC and HPH having the strongest effects. Compared with the control, protein retention in MF retentate from pre-treated samples decreased by 7.5–11.5%, with HPH + TSC showing the greatest effect, while permeate protein content increased by 5.45–9.64% ($P < 0.05$). SDS-PAGE confirmed lower levels of protein aggregation, particularly in HPH + TSC, coinciding with the lowest sedimented protein level (43.4%). Trisodium citrate pre-treated samples showed significantly lower ($P < 0.05$) calcium and magnesium levels, providing evidence that cations are involved in mediating protein aggregation. The results indicate that WPC treatment can modify protein permeation, improving the yield of WPI while also generating an MF retentate further enriched in polar lipids, supporting more sustainable dairy processing. © 2025 The Author(s). International Journal of Dairy Technology published by John Wiley & Sons Ltd on behalf of Society of Dairy Technology.

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Funding Source: Competitively-Won Funding

Research Programme: CareerFIT

Heat-induced protein aggregation influences protein partitioning during microfiltration of whey in the production of whey protein isolate

Mestawet A.T.; France T.C.; Mulcahy P.G.J.; O'Mahony J.A.

International Dairy Journal, 2026

This study was designed to investigate the effects of heat treatment and selective thiol blocking using N-ethylmaleimide (NEM) on protein partitioning during microfiltration (MF) of whey. Clarified sweet whey was subjected to five treatments: untreated control (CTRL), pH-adjusted (CTRLpH), pH + NEM (TNEM), pH + heat (THeat), and pH + NEM + heat (TNEM + Heat). Significant differences ($P < 0.05$) were observed in protein permeation and retention. Compared to CTRL, protein content in the permeate was 22 % and 16 % lower in THeat and TNEM + Heat, respectively. Conversely, protein retention in the retentate was 22 % and 16 % higher in the same treatments. These results confirm that disulphide bond-mediated aggregation reduces protein permeation during MF. The findings provide new insight into how thiol reactivity and thermal treatment influence protein transmission. This scientific insight supports the development of targeted strategies (e.g., reduced- or non-thermal processing) to increase protein yield and enhance processing sustainability in the production of value-added whey streams and whey protein ingredients. © 2026 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY license. <http://creativecommons.org/licenses/by/4.0/>

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Competitively-Won Funding Publications: REFLOW

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Research Programme: REFLOW

Struvite production from dairy processing wastewater: Optimizing reaction conditions and effects of foreign ions through multi-response experimental models

Numviyimana C.; Warchoń J.; Izydorczyk G.; Baśladyńska S.; Chojnacka K.

Journal of the Taiwan Institute of Chemical Engineers, 2020

Struvite is the preferred form of phosphorus recovery for fertilizer by chemical precipitation. The concentration of phosphorus in raw wastewater from dairy processing is higher than acceptable values for prevention of water pollution. Along with phosphorus, potassium and calcium are its main counterions with high concentration. Thus, calcium phosphate salts are prompt to precipitate and decrease struvite production. The effect of such phosphate counter-ions were optimized using design of experiments and desirability function to maximize both phosphorus recovery and struvite production. Under optimum conditions, the yields were 98.6 ± 1.1 and 85.7 ± 2.5 percent for phosphorus recovery and struvite precipitation, respectively. Factors optimization was achieved with desirability $D = 0.995$. By in-vitro assay of nutrients release, the product demonstrated better phosphorus availability than the one obtained with high calcium dose in reactor. The obtained molar ratios of dose can serve in wastewater treatment coupled to phosphorus precipitation with a fertilizer value product. © 2020 The Authors

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Research Programme: REFLOW

Systematic Review of Dairy Processing Sludge and Secondary STRUBIAS Products Used in Agriculture

Hu Y.; Khomenko O.; Shi W.; Velasco-Sánchez Á.; Ashekuzzaman S.M.; Bennegadi-Laurent N.; Daly K.; Fenton O.; Healy M.G.; Leahy J.J.; Sørensen P.; Sommer S.G.; Taghizadeh-Toosi A.; Trinsoutrot-Gattin I.

Frontiers in Sustainable Food Systems, 2021

Worldwide dairy processing plants produce high volumes of dairy processing sludge (DPS), which can be converted into secondary derivatives such as struvite, biochar and ash (collectively termed STRUBIAS). All of these products have high fertilizer equivalent values (FEV), but future certification as phosphorus (P)-fertilizers in the European Union will mean they need to adhere to new technical regulations for fertilizing materials i.e., content limits pertaining to heavy metals (Cd, Cu, Hg, Ni, Pb, and Zn), synthetic organic compounds and pathogens. This systematic review presents the current state of knowledge about these bio-based fertilizers and identifies knowledge gaps. In addition, a review and calculation of greenhouse gas emissions from a range of concept dairy sludge management and production systems for STRUBIAS products [i.e., biochar from pyrolysis and hydrochar from hydrothermal carbonization (HTC)] is presented. Results from the initial review showed that DPS composition depends on product type and treatment processes at a given processing plant, which leads to varied nutrient, heavy metal and carbon contents. These products are all typically high in nutrients and carbon, but low in heavy metals. Further work needs to concentrate on examining their pathogenic microorganism and emerging contaminant contents, in addition to conducting an economic assessment of production and end-user costs related to chemical fertilizer equivalents. With respect to STRUBIAS products, contaminants not present in the raw DPS may need further treatment before being land applied in agriculture e.g., heated producing ashes, hydrochar, or biochar. An examination of these products from an environmental perspective shows that their water quality footprint could be minimized using application rates based on P incorporation of these products into nutrient management planning and application by incorporation into the soil. Results from the concept system showed that elimination of methane emissions was possible, along with a reduction in nitrous oxide. Less carbon (C) is transferred to agricultural fields where DPS is processed into biochar and hydrochar, but due to high recalcitrance, the C in this form is retained much longer in the soil, and therefore STRUBIAS products represent a more stable and long-term option to increase soil C stocks and sequestration. Copyright © 2021 Hu, Khomenko, Shi, Velasco-Sánchez, Ashekuzzaman, Bennegadi-Laurent, Daly, Fenton, Healy, Leahy, Sørensen, Sommer, Taghizadeh-Toosi and Trinsoutrot-Gattin.

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Research Programme: REFLOW

Sewage Sludge Thermal Treatment Technologies with a Focus on Phosphorus Recovery: A Review

Kwapinski W.; Kolinovic I.; Leahy J.J.

Waste and Biomass Valorization, 2021

Phosphorus presents a limited, irreplaceable and essential nutrient necessary for the growth of organisms. There is an increasing effort to recover phosphorus from production waste streams. Sewage sludge presents an important source of phosphorus but also contains organic pollutants and heavy metals. Thermal treatment technologies seem to be a promising option to treat sewage sludge and obtain ash/char from which high recovery rate of phosphorus can be reached. In this review, sewage sludge management options in compliance with EU legal requirements are first reviewed. Follows, an overview of sewage sludge thermal treatment technologies including incineration, hydrothermal carbonisation, pyrolysis and gasification, for the purpose of phosphorus recapture. We summarize recent advances in thermal treatment processes of sewage sludge and phosphorus recovery, identify challenges and knowledge gaps. Thermochemical methods proved to have many advantages over pure wet chemical methods for phosphorus recovery. The review provides the foundation for future research aimed at achieving efficient, economic and environmental sustainable recapture of phosphorus from sludge thermal treatment products. Graphic Abstract: [Figure not available: see fulltext.]. © 2021, Springer Nature B.V.

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Research Programme: REFLOW

Nutrients recovery from dairy wastewater by struvite precipitation combined with ammonium sorption on clinoptilolite

Numviyimana C.; Warchoń J.; Ligas B.; Chojnacka K.

Materials, 2021

Struvite precipitation from Wastewater involves an excess of ammonium to create a supersaturated initial solution. The remaining fraction can be a threat to the environment. This work combined struvite precipitation and ammonium sorption using natural zeolite to decrease the ammonium level in the effluent. Two approaches of estimation of feed sample doses were used. One consisted of gradient experiments for ammonium precipitation to the asymptotic level and was combined with clinoptilolite to lower the ammonium level in the effluent. This approach used doses of 0.05:1.51:0.61:1 of Ca: Mg:NH₄⁺:PO₄³⁻ mole ratios, respectively. In contrast, three level design with narrowed NH₄⁺:PO₄³⁻ range reached 0.25:1.51:0.8:1 for Ca:Mg: NH₄⁺:PO₄³⁻ mole ratios. The addition of zeolite decreased effluent ammonium concentration. In both ways, the P and N recoveries were higher than 94% and 72%, respectively. The complexity of the precipitation mixture decreased the ammonium sorption capacity (Q_e) of clinoptilolite from Q_e of 0.52 to 0.10 meq·g⁻¹ in single and complex solutions, respectively. Thermodynamically, the addition of 1.5 % of clinoptilolite changed the struvite precipitation spontaneity from ΔG of -5.87 to -5.42 kJ·mol⁻¹ and from 9.66 to 9.56 kJ·mol⁻¹ for gradient and three level experimental procedures, respectively. Thus, clinoptilolite demonstrated a positive effect on the struvite precipitation process and its environmental impact. © 2021 by the authors. Licensee MDPI, Basel, Switzerland.

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Research Programme: REFLOW

Dairy processing sludge and co-products: A review of present and future re-use pathways in agriculture

Shi W.; Healy M.G.; Ashekuzzaman S.M.; Daly K.; Leahy J.J.; Fenton O.

Journal of Cleaner Production, 2021

The dairy industry is one of the largest global producers of wastewater and generates huge volumes of dairy processing sludge (DPS). There are two main types of DPS, lime-treated dissolved air floatation sludge and bio-chemically-treated activated sludge. These sludge types may also be converted to STRUBIAS (STRUvite, Blochar, AShes) products which have potential as fertilizers, secondary feedstocks for phosphate fertiliser granules, and soil amendments. A small number of studies indicate that these products have variable nutrient and metal contents, which differ across sludge and STRUBIAS product types. This is due to many factors such as the type of dairy plants, wastewater treatment process and production technologies. Although such products are land applied, the phosphorus (P) and nitrogen (N) fertilizer equivalency value (FEV) are often unknown and not factored into application rates, and therefore need study under field conditions (across soil and crop types). This review identifies a need to quantify antimicrobial drugs, hormones, pesticides, disinfectants, persistent organic pollutants (POPs), microplastics and nano-particles in all DPS and STRUBIAS types. Where detected, testing should follow the transfer of these contaminants to the soil, crop and water continuum. Further knowledge in the areas identified would enable both agronomic and environmental goals to be met and promote higher uptake of DPS and STRUBIAS re-use in agriculture. © 2021 The Author(s)

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Research Programme: REFLOW

A meta-analysis of LCAs for environmental assessment of a conceptual system: Phosphorus recovery from dairy wastewater

Behjat M.; Svanström M.; Peters G.

Journal of Cleaner Production, 2022

A significant increase in phosphorus-rich dairy wastewater coincides with a decrease in the availability of fossil phosphate rock resources in Europe. This confluence of events has led to the development of technologies for phosphorus recovery from dairy wastewater. This study aims to inform and guide such development with regard to life cycle environmental impacts prior to their implementation in dairy contexts. With the lack of inventory data at this point and the non-existence of earlier life cycle assessments on the use of phosphorus recovery technologies in a dairy context in literature, we performed a meta-analysis where we extracted and compared published results on life cycle environmental impacts from two fields (1) dairy industries, with a focus on the dairy wastewater treatment and (2) phosphorus recovery technologies in a municipal wastewater treatment context. The results show that despite its intended effect, normal dairy wastewater treatment in many cases still contributes significantly to eutrophication. Most of the phosphorus recovery technologies examined here exhibited a lower global warming potential and cumulative energy demand than those of dairy wastewater treatment processes. It indicates that problem shifting could be avoided when phosphorus recovery is introduced. However, no technologies involving incineration have had the impact of acidification reported which represents a potential knowledge gap since impacts are expected related to incineration emissions. A comparison between the extracted data for phosphorus recovery technologies shows that there are lower impacts related to technologies that recover phosphorus from the liquid phase, than from sludge or ash. © 2022 The Authors

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Research Programme: REFLOW

Hydrothermal carbonization (HTC) of dairy waste: effect of temperature and initial acidity on the composition and quality of solid and liquid products

Khalaf N.; Shi W.; Fenton O.; Kwapinski W.; Leahy J.J.

Open Research Europe, 2022

Background: Hydrothermal carbonization (HTC) of dairy processing waste was performed to investigate the effect of temperature and initial pH on the yield and composition of the solid (hydrochar) and liquor produced. All hydrochars met the EU requirements of organo-mineral solid fertilizers defined in the Fertilizing Products Regulation in terms of phosphorus (P) and mineral content. Methods: Laboratory scale HTC was performed using pressurized reactors, and the products (solid and liquid) were collected, stored and analyzed for elemental composition and nutrient content using Inductively coupled plasma optical emission spectroscopy (ICP-OES), ultraviolet-visible spectrophotometry (UV-Vis) and other analytic techniques. Results: Maximum hydrochar yield (60.67%) was observed at T=180°C and pH=2.25, whereas the maximum P-recovery was 80.38% at T=220°C and pH=4.6. The heavy metal content of the hydrochars was mostly compliant with EU limitations, except for Ni at T=220°C and pH=8.32. Meanwhile, further study of Chromium (Cr) species is essential to assess the fertilizer quality of the hydrochars. For the liquid product, the increase in temperature beyond 200°C, coupled with an increase in initial acidity (pH=2.25) drove P into the liquor. Simultaneously, increasing HTC temperature and acidity increased the concentration of NO₃⁻ and NH₄⁺ in the liquid products to a maximum of 278 and 148 mg/L, respectively, at T=180°C and pH=4.6. Furthermore, no direct relation between final pH of liquor and NH₄⁺ concentration was observed. Conclusions: HTC improved the nutrient content of dairy waste, allowing for the production of potential solid organo-mineral fertilizers requiring additional treatment to ensure safe fertilizer application. Copyright: © 2022 Khalaf N et al.

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Research Programme: REFLOW

Phosphorus recovery as struvite from hydrothermal carbonization liquor of chemically produced dairy sludge by extraction and precipitation

Numviyimana C.; Warchot J.; Khalaf N.; Leahy J.J.; Chojnacka K.

Journal of Environmental Chemical Engineering, 2022

Phosphorus (P) recovery from dairy wastewater involves its accumulation into phosphorus-rich sludge using a physico-chemical or biological process. The high iron content in chemical sludge decreases its usability in agricultural activities. The hydrothermal carbonization (HTC) is an option used to treat the sludge to obtain hydrochar for various applications, including its use as an energy source and as a carbon-dense material. The HTC process leaves a bigger volume of nutrient-rich liquor, which phosphorus (P) purification was the subject of this work. By direct precipitation, the product iron content was 17.96%, a value higher than accepted limits for phosphate fertilizers. Thus, P extraction followed by struvite precipitation was studied. The use of oxalic acid extracted 86.7% of P from HTC liquor, while 86.6% of iron was removed. The process conditions of pH 9, and salt dosage of 1.73:1.14:1 for Mg:NH₄⁺:P mole ratio for struvite precipitation were obtained with a P recovery of 99.96%, and the effluent P concentration below 2 mg·L⁻¹. The quality of products as fertilizers was tested by both in-vitro and in-vivo assays. High iron content in the product demonstrated a negative effect on plant germination, whilst the precipitation product from P extract demonstrated an advantage of P purification into struvite for plant macro and micronutrient availability. The used method of P extraction followed by struvite precipitation is useful for both P and iron recovery into two separate products with agricultural and chemical applications, respectively. © 2021 The Authors

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Research Programme: REFLOW

An examination of maximum legal application rates of dairy processing and associated STRUBIAS fertilising products in agriculture

Shi W.; Fenton O.; Ashekuzzaman S.M.; Daly K.; Leahy J.J.; Khalaf N.; Hu Y.; Chojnacka K.; Numviyimana C.; Healy M.G.

Journal of Environmental Management, 2022

The dairy industry produces vast quantities of dairy processing sludge (DPS), which can be processed further to develop second generation products such as struvite, biochars and ashes (collectively known as STRUBIAS). These bio-based fertilizers have heterogeneous nutrient and metal contents, resulting in a range of possible application rates. To avoid nutrient losses to water or bioaccumulation of metals in soil or crops, it is important that rates applied to land are safe and adhere to the maximum legal application rates similar to inorganic fertilizers. This study collected and analysed nutrient and metal content of all major DPS (n = 84) and DPS-derived STRUBIAS products (n = 10), and created an application calculator in MS Excel™ to provide guidance on maximum legal application rates for ryegrass and spring wheat across plant available phosphorus (P) deficient soil to P-excess soil. The sample analysis showed that raw DPS and DPS-derived STRUBIAS have high P contents ranging from 10.1 to 122 g kg⁻¹. Nitrogen (N) in DPS was high, whereas N concentrations decreased in thermo-chemical STRUBIAS products (chars and ash) due to the high temperatures used in their formation. The heavy metal content of DPS and DPS-derived STRUBIAS was significantly lower than the EU imposed limits. Using the calculator, application rates of DPS and DPS-derived STRUBIAS materials (dry weight) ranged from 0 to 4.0 tonnes ha⁻¹ y⁻¹ for ryegrass and 0–4.5 tonnes ha⁻¹ y⁻¹ for spring wheat. The estimated heavy metal ingestion to soil annually by the application of the DPS and DPS-derived STRUBIAS products was lower than the EU guideline on soil metal accumulation. The calculator is adaptable for any bio-based fertilizer, soil and crop type, and future work should continue to characterise and incorporate new DPS and DPS-derived STRUBIAS products into the database presented in this paper. In addition, safe application rates pertaining to other regulated pollutants or emerging contaminants that may be identified in these products should be included. The fertilizer replacement value of these products, taken from long-term field studies, should be factored into application rates. © 2021 The Authors

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Research Programme: REFLOW

Mineral fertiliser equivalent value of dairy processing sludge and derived biochar using ryegrass (*Lolium perenne* L.) and spring wheat (*Triticum aestivum*)

Shi W.; Healy M.G.; Ashekuzzaman S.M.; Daly K.; Fenton O.

Journal of Environmental Management, 2022

As supply chains of chemical fertilisers become more precarious, raw or derived bio-based fertilisers (herein referred to as bio-fertilisers) from the dairy processing industry could be good alternatives. However, their agronomic performance is relatively unknown, and where documented, the method to estimate this value is rarely presented. This pot study investigated aluminium-precipitated and calcium-precipitated dairy processing sludges (Al and Ca-DPS) and DPS-derived biochar as potential bio-fertilisers to grow ryegrass (*Lolium perenne* L.) and spring wheat (*Triticum aestivum*). The study aims were to examine how (1) application rate (optimal versus high) and (2) calculation methods (with and without chemical fertiliser response curves) can affect estimates of nitrogen and phosphorus mineral fertiliser equivalence value (N- and P-MFE) and associated agronomic advice. The results from both crops showed that for nitrogen application rates (125 or 160 kg ha⁻¹ for ryegrass and 160 or 240 kg ha⁻¹ for spring wheat) estimates of N-MFE increased for both Al-DPS and Ca-DPS as application rate increased. Dry matter yield response curves produced the highest % N-MFE results (e.g., ryegrass ~50% and 70% for Al-DPS and Ca-DPS) with other calculation methods producing all similar results (e.g., ryegrass ~20% for Al-DPS and Ca-DPS). For phosphorus application rates (40 or 80 kg ha⁻¹ for ryegrass and 50 or 80 kg ha⁻¹ for spring wheat), estimates of P-MFE did not increase with application rate. Negative P-MFE values obtained for Ca-DPS and DPS-biochar when growing ryegrass and spring wheat grain, respectively, indicated low plant available phosphorus. Overall, Al-DPS had better performance as a bio-fertiliser when compared to the other products tested. There was no significant difference between the two calculation methods of MFE, which suggests that the determination of MFE could be simplified by using one application as opposed to numerous application rates of fertilisers. Future work should focus on elucidating the N- and P-MFE of a wider range of DPS and STRUBIAS bio-fertilisers, and alternative methods should be investigated that enable a comparison across all bio-fertiliser types. © 2022

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Research Programme: REFLOW

Innovative multiple resource recovery pathways from EBPR wastewater treatment–derived sludge

Binder P.M.; Frison N.; Guerra-Gorostegi N.; Hidayat I.; Paredes L.; Llenas L.; Blázquez E.; Mora M.; Ballottari M.; Cazzaniga S.; Fatone F.; Salas S.P.

Biomass Conversion and Biorefinery, 2023

Wastewater treatment–derived sludge is a growing concern. Environmental issues, rising sludge production rates, and stringent regulations create the necessity to seek for treatment and valorization alternatives. Sludge is a potential source of high-value materials which can be recovered and transformed into new products such as animal feeds; bioplastics; biofuels, biostimulants; or biobased fertilizers. Considering the current legal constraints hindering the use of certain waste streams, the objective of this work is to show the technical viability for obtaining multiple valuable products from sludge. The emphasis is placed on novel valorization pathways, such as microalgae and purple bacteria cultures growing over sludge. The obtained products are benchmarked against traditional methods for resource recovery such as direct land application and P recovery from ashes. Our results show, besides the nutrient (TKN 7.38, TP 4.41; K 0.47 g 100 g TS-1) and energy content (HHV 22.53 MJ Kg-1 TS), that sludge could be employed to produce a suitable growing medium for microalgae and purple bacteria cultures obtaining, in the latter, remarkable high contents of high-quality proteins (64.50 % dw) for potential valorization as animal feed ingredient. We also obtained nutrient rich microalgae biomass (TKN 7.10, TP 8.10; K 0.40 g 100 g TS-1) which could be used as inputs for biobased fertilizers or biostimulants preliminarily complying with the nutrient requirements in EU 2019/1009. Current global scenario, showing economic and supply risk uncertainties regarding food production inputs, generates the urgent need to find feasible pathways for obtaining recovered products such as the ones presented in this study. Graphical abstract: [Figure not available: see fulltext.]. © 2023, The Author(s).

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Research Programme: REFLOW

A Novel Hybrid Membrane Process Coupled with Freeze Concentration for Phosphorus Recovery from Cheese Whey

Hidayat I.; Paredes L.; Binder P.M.; Guerra-Gorostegi N.; Mora M.; Ponsá S.; Oatley-Radcliffe D.L.; Llenas L.

Membranes, 2023

The ever-increasing demand for phosphorus fertilisers for securing global food production, coupled with finite phosphate rock reserves, is one of the emerging problems in the world. Indeed, phosphate rock is listed as an EU critical raw material, triggering attention to find an alternative source to substitute the use of this limited resource. Cheese whey, characterized by a high content of organic matter and phosphorus, represents a promising feedstock for phosphorus recovery and recycling. An innovative application of a membrane system coupled with freeze concentration was assessed to recover phosphorus from cheese whey. The performances of a microfiltration membrane (0.2 µm) and an ultrafiltration (200 kDa) membrane were evaluated and optimized under different transmembrane pressures and crossflow velocities. Once the optimal operating conditions were determined, a pre-treatment including lactic acid acidification and centrifugation was applied to increase the permeate recovery. Finally, the efficiency of progressive freeze concentration for the treatment of the permeate obtained from the optimum conditions (UF 200 kDa with TMP of 3 bar, CFV of 1 m/s and lactic acid acidification) was evaluated at specific operating conditions (-5 °C and 600 rpm of stirring speed). Finally, 70% of phosphorus could be recovered from cheese whey using the coupled technology of the membrane system and freeze concentration. A phosphorus-rich product was obtained with high agronomic value, which constitutes a further step towards establishing a broader circular economy framework. © 2023 by the authors.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Effects of dairy processing sludge and derived biochar on greenhouse gas emissions from Danish and Irish soils

Hu Y.; Thomsen T.P.; Fenton O.; Sommer S.G.; Shi W.; Cui W.

Environmental Research, 2023

Globally, to ensure food security bio-based fertilizers must replace a percentage of chemical fertilizers. Such replacement must be deemed sustainable from agronomic and greenhouse gas (GHG) emission perspectives. For agronomic performance several controlled protocols are in place but not for testing GHG emissions. Herein, a pre-screening tool is presented to examine GHG emissions from bio-waste as fertilizers. The various treatments examined are as follows: soil with added mineral nitrogen (N, 140 kg N ha⁻¹) fertilizer (MF), the same amount of MF combined with dairy processing sludge (DS), sludge-derived biochar produced at 450 °C (BC450) and 700 °C (BC700) and untreated control (CK). These treatments were combined with Danish (sandy loam) or Irish (clay loam) soils, with carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O) emissions and soil inorganic-N contents measured on selected days. During the incubation, biochar mitigated N₂O emissions by regulating denitrification. BC450 reduced N₂O emissions from Danish soil by 95.5% and BC700 by 97.7% compared to emissions with the sludge application, and for Irish soil, the N₂O reductions were 93.6% and 32.3%, respectively. For both soils, biochar reduced CO₂ emissions by 50% as compared to the sludge. The lower N₂O reduction potential of BC700 for Irish soil could be due to the high soil organic carbon and clay content and pyrolysis temperature. For the same reasons emissions of N₂O and CO₂ from Irish soil were significantly higher than from Danish soil. The temporal variation in N₂O emissions was correlated with soil inorganic-N contents. The CH₄ emissions across treatments were not significantly different. This study developed a simple and cost-effective pre-screening method to evaluate the GHG emission potential of new bio-waste before its field application and guide the development of national emission inventories, towards achieving the goals of circular economy and the European Green Deal. © 2022

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Phosphorus recovery from hydrothermal carbonization of organic waste: a review

Khalaf N.; Leahy J.J.; Kwapinski W.

Journal of Chemical Technology and Biotechnology, 2023

Background: This review sheds light on the topic of phosphorus (P) recovery from hydrothermal carbonization (HTC) of organic waste. The paper focuses on the rising need for securing alternative P sources for the increasing demand, in addition to the development of HTC as a thermochemical technique for waste valorization. **Results:** This article studies the definition of P-recovery from HTC, while introducing the notion of availability, which is a significant factor for studying the efficiency of P-recovery. A brief comparison between different treatment techniques for P-recovery from waste streams is presented, and the advantages of HTC are highlighted among different thermochemical techniques. Furthermore, the mechanisms of P-transformation during HTC reactions are studied, and the effect of various parameters on P-destination and recovery is emphasized. Finally, applications of P-recovery from HTC products on the laboratory and industrial scales are studied to assess the feasibility of the application. **Conclusion:** In short, the paper offers a detailed insight into the definition, mechanism, and potential feasibility of P-recovery from HTC of organic wastes within the scopes of resource management, waste valorization, and fertilizer production. Even though HTC for P-recovery from organic waste has been applied on laboratory and industrial scales, several challenges persist towards optimizing this process. The paper concludes that adopting P-availability as a criterion for efficiency is necessary to optimize HTC conditions for P-recovery. Further studies on P-transformation during later-stage reactions of HTC are recommended, as well. © 2023 The Authors. Journal of Chemical Technology and Biotechnology published by John Wiley & Sons Ltd on behalf of Society of Chemical Industry (SCI). © 2023 The Authors. Journal of Chemical Technology and Biotechnology published by John Wiley & Sons Ltd on behalf of Society of Chemical Industry (SCI).

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Research Programme: REFLOW

A comparative study of thermally and chemically treated dairy waste: Impacts on soil phosphorus turnover and availability using ^{33}P isotope dilution

Khomenko O.; Fenton O.; Leahy J.J.; Daly K.

Journal of Environmental Management, 2023

Dairy processing sludge (DPS) and DPS-derived secondary products such as struvite, biochar, hydrochar and ash (collectively known as SRUBIAS) are emerging as alternatives to fertilizers produced from mined rock phosphate. However, little is known about how these products affect soil P availability and daily P turnover rates. A lack of such information prevents precision nutrient management planning using these products out on farms. This study used a novel isotope dilution technique (IPD) with ^{33}P as a tracer to compare P turnover in soils amended with chemically (alum-treated DPS and struvite) and thermally (biochar, hydrochar, ash) treated DPS. Results showed that thermally treated products exhibited poor agronomic performance as P fertilizers, potentially inhibiting P availability when applied to soils. For example, a P deficient soil amended with hydrochar treatment at the highest application rates did not record a build-up of available P to agronomic target values. In ash and biochar treated P deficient soils, available P increased but only with very high application rates of 150 and 80 mg P kg⁻¹. The application of these products as fertilizers could have negative implications for both environmental and agronomic goals. Conversely, chemically treated fertilisers demonstrated better agronomic performance. The same agronomic target value was reached with application rates of only 20 mg P kg⁻¹ soil for DPS and 50 mg P kg⁻¹ soil for struvite. However, the techniques deployed revealed that these products exhibited slower rates of available and exchangeable P build-up when compared with chemical fertilisers. This suggests that these bio-based alternatives require higher application rates or earlier application times compared to conventional chemical fertilizers. Regulations providing advice on P use in agricultural soils need to account for slower P turnover in soils receiving recycled fertilizers. The IPD technique is transferrable to all wastes to examine their performance as fertilizers. © 2022 The Authors

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Research Programme: REFLOW

Changes in phosphorus turnover when soils under long-term P management are amended with bio-based fertiliser

Khomenko O.; Fenton O.; Leahy J.J.; Daly K.

Geoderma, 2023

Understanding available phosphorus (P) turnover could improve sustainable P management. An isotope tracing ^{33}P was used to measure daily P turnover rates and exchangeable P (E) in P deficient, balanced, and excess P soils from a long-term P site. Turnover under P deficient conditions was characterised by the lowest P flux predominantly from the available into the exchangeable P pools (P efflux). The P efflux in the P deficient soil was $3.7 \pm 0.6 \text{ mg P/L day}^{-1}$ and the inverse flux (P influx) was $1.6 \pm 0.4 \text{ mg P/L day}^{-1}$. Turnover rates were more than twofold higher under P balanced and surplus conditions, exhibiting an equilibrium between influx and efflux rates. The contribution of abiotic processes to P turnover was predominant in the excess P soil, whereas biotic processes dominated turnover rates under P deficient conditions. Changes in P turnover were measured following application of single superphosphate (SSP) and dairy processing sludge (DPS). Both fertiliser types increased P turnover rates and availability across all soils. After SSP application, E values plateaued between 238 and 297 mg P/L regardless of initial P status. Slower P release from DPS was evidenced by a wider range of E values (97–160 mg P/L) with slower turnover. © 2022

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Research Programme: REFLOW

Optical chemical sensors for soil analysis: possibilities and challenges of visualising NH₃ concentrations as well as pH and O₂ microscale heterogeneity

Merl T.; Hu Y.; Pedersen J.; Zieger S.E.; Bornø M.L.; Tariq A.; Sommer S.G.; Koren K.

Environmental Science: Advances, 2023

Agricultural nitrogen (N) application to soils is the main source of atmospheric ammonia (NH₃). Ammonia negatively impacts the environment on a large scale. However, emissions of NH₃ are affected by spatiotemporal heterogeneities of soil parameters on a microscale. Some key parameters controlling processes of the N cycle are soil oxygen (O₂) and pH. To better understand biogeochemical soil processes, NH₃ emissions and the interconnection of the ecospheres, we propose the application of optical chemical sensors (optodes) in and above soils. The use of optodes in soil science is in its infancy. In this laboratory-based study, we investigated the possibilities and challenges of using optodes in non-waterlogged soils with the extended application of a recently developed NH₃ optode along with pH and O₂ optodes in two different soils and with different fertilisers. Our intention is to help expand the use of optodes in soil science. Our results demonstrated the possibility to visualise reductions of NH₃ concentrations by 76% and 87% from the incorporation of sludge compared to the surface application of sludge. We showed from 2D measurements how soil pH and fertiliser composition correlate with NH₃ volatilisation. Our measurements demonstrated that pH optodes can have advantages over conventional methods when measuring pH in soils in situ but are challenged by the limited dynamic range (typically 3 pH units) compared to pH electrodes. Finally, we investigated the spatiotemporal dynamics of O₂ at different soil water contents and discuss potential challenges, which can lead to measuring artifacts. © 2023 RSC.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Assessment of willingness-to-pay for bio-based fertilisers among farmers and agricultural advisors in the EU

Moshkin E.; Garmendia Lemus S.; Bamelis L.; Buysse J.

Journal of Cleaner Production, 2023

Modern agricultural systems heavily depend on replenishing nutrients in the soils via mineral fertilisers. However, the extensive use and production of mineral fertilisers lead to adverse environmental effects. Furthermore, the raw materials used for the production of mineral fertilisers are unevenly distributed in the world and are susceptible to price fluctuations on the international market. A more circular solution is needed to ensure the sustainable supply of nutrients for agriculture in Europe. Bio-based fertilisers (i.e. fertilisers recycled from various waste streams while avoiding the use of fossil resources) can be a solution. The transition requires significant marketing efforts, but there is very little information on pricing preferences for newly developed bio-based fertilisers. This article aims to fill the gap by performing analyses of willingness-to-pay and price sensitivity using the Van Westendorp pricing technique and its extended version. Our analyses exemplified how the Van Westendorp methodologies can be used to generate insights into the price sensitivity of farmers and agricultural advisors in the EU. The selected methodologies allowed us to consider the ‘revenue vs. market share’ trade-off and thus distinguish the prices that are needed to gain the largest product market share and the prices that are needed to maximise the revenue from the products. Our results suggest that the price for a bio-based fertiliser at the 30–46% discount compared to the price of an equivalent mineral fertiliser would allow to maximise the market share of the product. Yet, to maximise the revenue the prices can be set equivalent to the ones of the mineral fertilisers. Combined with benchmarking studies and technical economic assessments our results contribute to the understanding of the key aspects needed for the development of marketing strategies and business case analyses for newly introduced products, such as bio-based fertilisers. © 2023

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Research Programme: REFLOW

Phosphorus fertiliser equivalent value of dairy processing sludge-derived STRUBIAS products using ryegrass (*Lolium perenne* L.) and spring wheat (*Triticum aestivum*)

Shi W.; Fenton O.; Ashekuzzaman S.M.; Daly K.; Leahy J.J.; Khalaf N.; Chojnacka K.; Numviyimana C.; Warchot J.; Healy M.G.

Journal of Plant Nutrition and Soil Science, 2023

Background: Struvite, biochar and ash products (collectively known as STRUBIAS) derived from different waste streams are used as fertilisers in agriculture. Raw dairy processing sludge (DPS) shows promise as bio-based fertilisers, but secondary STRUBIAS-derived products need further testing as fertilisers. Aims: The objective of this study was to calculate the phosphorus mineral fertiliser equivalency (P-MFE) for some STRUBIAS products derived from DPS. Methods: Ryegrass (*Lolium perenne* L.) and wheat (*Triticum aestivum*) pot trials were used to determine the P-MFE using the apparent P recovery (APR) method for Fe-DPS and DPS-derived struvites (Struvite 1–4), hydrochars (HC1–3) and ash. Results: The tested STRUBIAS products can be divided into two groups: (1) a range of products that can (i.e. Struvite 1–3) and (2) cannot (i.e., Struvite 4, HC1–3, ash and Fe-DPS) be considered fertilisers. In the first group, the P-MFE ranged from 66.8% to 76.7% for ryegrass and from 77.9% to 93.5% for spring wheat grain. In the second group, the P-MFE ranged from 7.8% to 58.3% for ryegrass and from –34.5% to –151.3% for spring wheat grain. The negative agronomic effects of some products for wheat grain (struvite and HC) in this study were mainly caused by high Fe content, which could be overcome by improved treatment processes. Conclusions: Future policy and research must be aware that not all the DPS-derived STRUBIAS products are suitable as fertilisers and therefore need to be tested individually. © 2023 The Authors. Journal of Plant Nutrition and Soil Science published by Wiley-VCH GmbH.

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Research Programme: REFLOW

Environmental assessment of phosphorus recovery from dairy sludge: A comparative LCA study

Behjat, M., Svanstrom, M. and Peters, G.

Waste Management, 2024

Phosphate rock is a finite, non-renewable mineral resource that is used primarily in fertiliser production. The scarcity and the increasing demand for this finite material led the European Commission to include it in the critical raw material list in 2014. As a consequence, efforts have been directed towards enhancing material use efficiency, initiating recycling efforts, and formulating waste policies to mitigate the criticality of raw materials. Interest in the development of technologies for nutrient recovery from organic waste streams has increased in recent years, and dairy processing sludge (DPS) is a potential input waste stream. Although the recovery of P from DPS can contribute to more circular flows of nutrients in society, it has to be assessed whether there are also overall environmental gains. This paper reports on a life cycle assessment (LCA) of the environmental impacts of three scenarios for phosphorus (P) recovery involving hydrothermal carbonization (HTC) and struvite precipitation and a comparison to a reference drying scenario. HTC produces a solid fraction (hydrochar), and a liquid fraction (process water) and in one of the scenarios (Scenario 3), leaching the hydrochar for additional P recovery is considered. From the process water as well as from the hydrochar leachate, P is precipitated in the form of struvite. Scenarios 1 and 2 both consider HTC and struvite production with the only difference that the hydrochar is used as a fuel instead of as a fertilizer in the latter case, and Scenario 3 adds leaching of the hydrochar with subsequent struvite production and considers that hydrochar is used as a fuel. In the fourth (reference) scenario, dewatering and drying of DPS is considered. The recovered product use in agriculture was not assessed at this stage. The assessment of the emerging technologies in Scenarios 1–3 was done by studying the technologies in early stages of development but modelling them as more developed in the future. Additional functions beyond the functional unit of one kg of P recovered were handled through a system expansion by substitution approach. This way, the system was credited for calcium ammonium nitrate (CAN) production in all scenarios and for wood chips production in Scenarios 2 and 3. Looking at net outcomes for all scenarios, the life cycle impact indicator results for scenario 2 are lower than the other scenarios in several impact categories. Large gains in scenario 2 are related to the avoided production of wood chips.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Sustainability indicator identification and selection for an innovative conceptual system: Phosphorus recovery from dairy wastewater

Behjat, M., Svanstrom, M.; Peters, G.; Perez-Soba, M.

Resources, Conservation and Recycling, 2024

In Europe, a decrease in the availability of phosphate rock resources has led to the development of emerging technologies for phosphorus recovery, with the purpose of generating products that can be used as fertilisers. An innovative conceptual system dedicated to the phosphorus recovery from dairy wastewater is considered in the paper. New technologies need to be assessed using relevant sustainability indicators. In this study, we developed an approach for identifying and selecting indicators. Based on searches of literature and expert interviews, three different tools were developed: an indicator screening framework, a questionnaire for finding actor priorities, and a list of indicator selection criteria. The new approach was successfully used to narrow down an initial set of 382 indicators identified in the literature to 26 that were considered representative and practicable for the assessment of the considered system.

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Research Programme: REFLOW

European farmers' perceptions and intentions to use bio-based fertilisers: Insights from the theory of planned behaviour and perceived utility

Garmendia-Lemus S.; Moshkin E.; Hung Y.; Tack J.; Buysse J.

Journal of Cleaner Production, 2024

Bio-based fertilisers, derived from diverse biological waste and processing technologies, have shown promising efficiency. However, the successful introduction of newly developed bio-based products in the market remains a challenge due to insufficient information on optimal marketing strategies. In this study, we examine farmers' perceptions regarding the acceptance of bio-based fertilisers in the EU. This research utilizes the Theory of Planned Behaviour (TPB) and an extended version with Perceived Utility as a mediating variable, using two distinct Structural Equation Models (SEMs) for analysis. Data were collected via an online questionnaire distributed across the EU 27 (n=332). Our findings reveal that farmers make rational decisions about fertiliser use, striving to maximize utility. Intentions to adopt bio-based fertilisers are strongly linked to attitudes, subjective norms, and perceptions of utility. Social networks and expert acceptance play a crucial role in shaping farmers' intentions. To enhance utility perception and encourage adoption, targeted marketing strategies should address key social groups, providing comprehensive information on the benefits and costs of implementation. Moreover, ensuring price stability through effective regulatory tools and mechanisms is vital for successful integration into the market. These insights offer valuable guidance to policymakers and the fertiliser industry, facilitating the introduction of bio-based fertilisers into the EU market. © 2023

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Research Programme: REFLOW

Hydrochar from dairy sludge as phosphorus fertiliser affects greenhouse gas emissions and maize yield

Hu Y., Taghizadeh-Toosi A., Baral K.R.; Smith A.M., Khalaf N., Sommer S.G.

Acta Agriculturae Scandinavica Section B: Soil and Plant Science, 2024

Dairy processing sludge is a phosphorus (P) rich waste with a high potential to replace mineral phosphorus fertiliser in crop production, with possible enhancement of greenhouse gas emissions to the environment. Hydrothermal carbonisation is a technology that transforms the sludge into a hydrochar. The objective of this study is examining P availability of two hydrochars produced from Danish and Irish dairy sludge and their influence on greenhouse gas emissions and maize yields. The trial assessed (i) Danish dairy sludge; (ii) hydrochar derived from Danish sludge; (iii) hydrochar made from Irish dairy sludge; (iv) mineral phosphorus fertiliser; and (v) control. Emissions of nitrous oxide and carbon dioxide, soil pH, mineral nitrogen contents and crop yields were measured. Treatment with Danish dairy sludge had significantly higher cumulative nitrous oxide emissions while the emissions from both hydrochars were not significantly different compared to mineral phosphorous feriliser. Statistical modelling showed that temperature, soil nitrate content, interactions both between temperature and precipitation, and between soil moisture and precipitation were drivers for nitrous oxide emissions. There was no difference in emissions among all treatments when scaled for yield. Hydrochar may alleviate the enhanced nitrous oxide emissions in soil without constraining P availability and maize crop yields. © 2024 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Tracing N₂O from dairy processing sludge amended soil with visualizing microscale heterogeneity of NH₃ and pH (Short Communication)

Hu, Y.; Merl, T.; Pedersen, J.; Bornø, ML.; Tariq, A.; Koren, K.; Sommer, S.G.

Atmospheric Environment: X, 2024

Nitrous oxide (N₂O) emissions from organic waste and animal slurry contribute to climate change and endanger our ecosystems. For the development of efficient mitigation technologies, in-depth knowledge of emission processes is needed. This can be obtained by non-destructive, temporal measurements of in-situ soil profiles and the transformation of ammonium (NH₄⁺) during events of emissions. Planar optode imaging is a non-destructive measuring method that can be used to visualize spatiotemporal changes of ammonia (NH₃) and pH in soil systems. In this study, soil amended with dairy processing sludge (DPS) was incubated in static chambers for 23 days, and GHG emissions, NH₃ concentrations and pH in the soil were measured simultaneously over time. The aim was to investigate the potential of applying different planar optodes to provide information that gives insight into processes of N₂O emissions. The DPS was applied to the soil as a surface layer (SL), with untreated soil as a control (CK). We were able to measure N₂O emissions while monitoring spatiotemporal changes of soil pH and NH₃ concentrations. The visualized microscale heterogeneity of the soil contributed to a better understanding of N₂O emission processes. While technical challenges (e.g., humidity sensitivity of the NH₃ optode and airtightness of the chambers) still need to be overcome, the method is a promising non-destructive method to study soil processes after application of different types of soil amendments.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Contrasting Phosphorus Build-up and Drawdown Dynamics in Soils Receiving Dairy Processing Sludge and Mineral Fertilisers

Khomenko O., Fenton O., Leahy J.J., Daly K.

Journal of Soil Science and Plant Nutrition, 2024

Sustainable utilisation of waste from the food industry is required to transition to a circular economy. The dairy industry relies on high phosphorus (P) inputs and produces large quantities of P-rich dairy processing sludge (DPS). Recycling DPS into P fertilisers provides an opportunity to decrease the reliance on chemical P fertilisers. However, current soil nutrient management planning (NMP) is based on chemical P and does not account for recycled alternatives. A pot trial using a novel isotope pool dilution technique was used to describe build-up and drawdown cycles of P in soils fertilised with DPS. Changes in available, exchangeable, and Mehlich3 P (M3-P) pools were recorded over 36 weeks of grass growth. Results demonstrated that in the period of high P demand (12 weeks), these P pools were depleted. As crop growth and demand decreased, available P recovered through mobilisation of P from exchangeable P and M3-P reserves. DPS allowed available P to recover and build up to agronomic target levels after 24 weeks. Using DPS, build-up of available and exchangeable P was slower but P use efficiency was higher at stages of slow growth. Dairy waste created a more stable P pool which could be utilised by crops over a growing season indicating that NMP needs to account for this in the decision support for growers. Isotope studies revealed that extractive agronomic tests do not capture drawdown in P reserves. © The Author(s) 2023.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Phosphorus Release Dynamics from Ashes during a Soil Incubation Study: Effect of Feedstock Characteristics and Combustion Conditions

Singla Just, B.; Binder, P.M.; Guerra-Gorostegi, N.; Diaz-Guerra, L.; Vilaplana, R.; Frison, N.; Meers, E.; Llenas, L.; Robles Aguilar, A.

Agronomy, 2024

Recovering phosphorus (P) through combustion from waste streams, like wastewater sludge and animal manure, offers a promising solution. This research explores the P release patterns in different ashes derived from secondary raw materials, using a long-term soil incubation lasting 160 days. The study evaluated the P release dynamics in five types of ashes from enhanced biological phosphorus removal (EBPR) systems and pig slurry burned at different temperatures. According to the results, a primary effect was observed on P bioavailability during the initial incubation period. All tested ashes release more than 50% of the total P applied between days 5 and 10. Ashes from EBPR exhibited higher P release than those from pig manure, indicating ash origin as a key factor in P release. Additionally, combustion temperature was crucial, with higher temperatures resulting in increased P release rates. Furthermore, the Pearson correlation revealed a strong relationship between the characteristics of the ashes and the amount of P release. Overall, these findings suggest that ashes could be a valuable P-source for agriculture avoiding the process of wet chemical P extraction, thus reducing both economic and environmental costs.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Do investments in phosphorus recovery from dairy processing wastewater pay off?

Uhlemann, J.-P.R.; Oude Lansink, A., Leahy, J.J., Dalhaus, T.

Journal of Environmental Management, 2024

While phosphorus fertilizers contribute to food security, part of the introduced phosphorus dissipates into water bodies leading to eutrophication. At the same time, conventional mineral phosphorus sources are increasingly scarce. Therefore, closing phosphorus cycles reduces pollution while decreasing trade dependence and increasing food security. A major part of the phosphorus loss occurs during food processing. In this article, we combine a systematic literature review with investment and efficiency analysis to investigate the financial feasibility of recovering phosphorus from dairy processing wastewater. This wastewater is particularly rich in phosphorus, but while recovery technologies are readily available, they are rarely adopted. We calculate the Net Present Value (NPV) of investing in phosphorus recycling technology for a representative European dairy processing company producing 100,000 tonnes of milk per year. We develop sensitivity scenarios and adjust the parameters accordingly. Applying struvite precipitation, the NPV can be positive in two scenarios. First, if the phosphorus price is high (1.51 million EUR) or second if phosphorus recovery is a substitute for mandatory waste disposal (1.48 million EUR). However, for a variety of methodological specifications, the NPV is negative, mainly because of high input costs for chemicals and energy. These trade-offs between off-setting pollution and reducing energy consumption imply, that policy makers and investors should consider the energy source for phosphorus recovery carefully.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Soil microorganisms increase Olsen phosphorus from poorly soluble organic phosphate: A soil incubation study

Velasco-Sánchez Á.; Bennegadi-Laurent N.; Trinsoutrot-Gattin I.; van Groenigen J.W.; Moinet G.Y.K.

Soil Use and Management, 2024

The potential shortage of mineral phosphorus (P) sources and the shift towards a circular economy motivates the introduction of new forms of P fertilizers in agriculture. However, the solubility of P in new fertilizers as well as their availability to plants may be low. In this experiment, we incubated an agricultural soil poor in P (28 mg P₂O₅ kg⁻¹) for 63 days in the presence of a range of organic and inorganic poorly soluble P forms commonly found in new fertilizers: hydroxyapatite (P-Ca), iron phosphate (P-Fe), phytic acid (P-Org) and a combination of P-Ca and P-Org (P-Mix). Cellulose and potassium nitrate (KNO₃) were added to stimulate microbial activity at the beginning of the incubation. We included a positive control with triple superphosphate (TSP) and negative controls with no P application (with and without cellulose and KNO₃). We assessed the fate of the different poorly soluble P forms in NaHCO₃ extracts (Olsen P) over time as a proxy for plant-available P. Soil microbial biomass, fungal to bacterial ratio, soil respiration, enzymatic activities (β -glucosidase, arylamidase and acid and alkaline phosphatase), N mineralization and soil pH were also monitored. At the beginning of the incubation, TSP showed the highest Olsen P across all treatments and P-Fe showed higher levels of Olsen P than the other poorly soluble P forms ($p < .05$). During the incubation, the levels of Olsen P decreased over time for TSP (positive control). Contrastingly, Olsen P increased significantly over time for all the poorly soluble P forms and the negative controls, indicating an increase in plant-available P. Particularly, levels of Olsen P for the P-Org treatment roughly doubled (shifting from 16.5 mg kg⁻¹ to 32.9 mg kg⁻¹) over the whole incubation period. The rate of increase in Olsen P was positively correlated with microbial biomass C:P ratio ($p < .01$) for all poorly soluble treatments. The higher levels of Olsen P for the P-Org treatment were also explained by a positive correlation with fungal biomass. Our results show that poorly soluble forms of P may be made available to plants under the influence of the microbial community, with a stronger effect on organic P forms. © 2023 The Authors. Soil Use and Management published by John Wiley & Sons Ltd on behalf of British Society of Soil Science.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Combination of *Lolium perenne* L. and *Festuca arundinacea* Schreb. improve yields under low phosphorus availability

Velasco-Sánchez, Á.; Ferron, L.M.E.; Mani, D.T.C.; Bennegadi-Laurent, N.; Trinsoutrot-Gattin, I.; Van Groenigen, J.W.; Moinet, G.Y.K.

Nutrient Cycling in Agroecosystems, 2024

Phosphorus (P) is one of the main nutrients for all plants, including grasses. However, sources of P fertilizer are not renewable, are not evenly distributed and overfertilization can lead to serious environmental degradation. Smart combinations of grasses may be able to more efficiently take up P from soils through complementarity. In a two-year field mesocosm experiment, we compared the performance of *Lolium perenne* L. and *Festuca arundinacea* Schreb. in monocultures and in combination, as well as a mixture of both species with a tetraploid variety of *Lolium perenne* L and *Phleum pratense* L. Plants were grown in an unfertilized low P soil and in P fertilized soil for two growing seasons. We measured biomass production, root traits, nutrient uptake, microbial biomass and enzymatic activities. In the unfertilized plots the combination of *Lolium perenne* and *Festuca arundinacea* generated the highest cumulative yields ($25,951 \pm 4059 \text{ kg ha}^{-1}$), relative total yield (> 1) and P nutrition index (0.79). We related this to the complementarity found in root traits and lower intraspecific competition of *Festuca arundinacea* and *Lolium perenne* diploid. *Festuca arundinacea* produced higher root biomass than *Lolium perenne* diploid at deeper soil layers (98 vs. 44 g m⁻²; $p < 0.05$). On the other hand, *Lolium perenne* diploid had significantly finer roots than *Festuca arundinacea* both at topsoil and bottom layers (0.19 vs. 0.22 mm and 0.19 vs. 0.23 mm at top and bottom layers respectively). The 4 species combination did not result in higher yields. Our results show that, in low P soils, combinations of grass species with contrasting root traits could lead to significantly higher yields than monocultures.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Phosphorus fertilization of grasses with dairy processing waste materials and contributions of soil microorganisms towards phosphorus uptake

Velasco-Sánchez Á.; Hu Y.; Sommers S.G.; Moinet G.Y.K.; Van Groenigen J.W.; Trinsoutrot-Gattin I.; Bennegadi-Laurent N.

Geoderma, 2025

Dairy processing waste (DPW) is one of the largest agro-industrial residues globally and has the potential of reducing the dependency on mineral phosphorus (P) fertilizers in grasslands. However, the agronomical value and the effects of DPW and the secondary materials produced from it on grass yields and soil microorganisms remains largely unexplored. In a 112-day greenhouse experiment, we evaluated the biomass production and P uptake of two grass species, grown on a loamy topsoil, with contrasting growth strategies (*Lolium perenne* L. fast-grower vs *Dactylis glomerata* L. slow-grower) and the effects on soil microorganisms under the fertilization of raw DPW sludge, and its secondary materials (ash and hydrochar). We compared the fertilized pots with a positive control with triple-superphosphate (TSP) and a negative control without P application. Our results showed that Olsen P and plant P concentrations were highest in the treatment with TSP after one month of application for both grass species. However, biomass production, P uptake and phosphorus use efficiency (PUE) were the greatest for sludge at day 112 for both species (56.39 ± 3.34 g pot⁻¹, 0.20 g \pm 0.02 P and 56.18 ± 12.44 %, n = 8). Ash and hydrochar resulted in lower biomass production than TSP and sludge and their PUE was low (< 20 %). Fungi and arbuscular mycorrhiza fungi PLFA markers were among the most important variables in explaining P uptake. The results of our study suggest that application of DPW sludge and soil's fungal biomass can reduce the dependency on mineral P fertilizers in grassland. © 2025 The Author(s)

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Phosphorus Fractionation of Dairy Processing Waste Recycled Fertilizers Reveals Inadequacy of the Standards, Measurement and Testing (SMT) Protocol

Velasco-Sánchez Á.; Khalaf N.; Leahy J.J.; Bennegadi-Laurent N.; Trinsoutrot-Gattin I.;
Van Groenigen J.W.; Moinet G.Y.K.

Communications in Soil Science and Plant Analysis, 2025

The Standards, Measurements and Testing (SMT) protocol is widely used to fractionate phosphorus (P); however, it lacks the focus on defining the solubility of P-fractions, which is usually performed independently. In this paper, the addition of a separate pre-wash step with H₂O as a first step prior to the SMT protocol was tested to account for the solubility of P. Results were compared to a control unmodified SMT experiment. The differences in P-fractions were analyzed to determine the origin of the readily available P (soluble P). Six different dairy processing waste treatment products were investigated in the form of sludge, hydrochar, and ash. Water-soluble P (WSP) was correlated with weakly bound calcium (Ca), aluminum (Al) and iron (Fe). However, the SMT protocol failed to correctly identify the different pools of P as unexpected correlations were found between P and Ca, Al and Fe. Moreover, large concentrations of organic P were present in ashes (>10 mg P g⁻¹). The organic P fraction included substantial amounts of Fe that correlated highly with P ($R^2 = 0.84$). No association between WSP and any of the pools of P defined by SMT was detected, with the exception of total P and inorganic P. The paper concludes that SMT erroneously classifies P into different discrete fractions across various recycled P-products with fertilizer potential. A critical reevaluation of the SMT protocol is recommended, by abandoning the categorization of P into discrete pools and switching to solvent-based categories referring to the chemicals used in each extraction. © 2025 The Author(s). Published with license by Taylor & Francis Group, LLC.

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Funding Source: Competitively-Won Funding

Research Programme: REFLOW

Hydrothermal carbonisation & struvite precipitation from dairy sludge: Evaluating market uncertainties & scale trade-offs

Garmendia-Lemus S.; Moshkin E.; Moloeznik-Paniagua D.; Khalaf N.; Numvimiyana C.;
Behjat M.; Tack J.; Huylensbroeck G.V.; Buysse J.

Waste Management, 2026

This study presents a techno-economic assessment of Hydrothermal Carbonisation (HTC) and Struvite Precipitation (STR) for dairy processing sludge (DPS), focusing on energy and cost performance across five system scales (2500–50,000 t/year). A dual approach was employed: a deterministic analysis using fixed input values, and a stochastic Monte Carlo simulation to assess uncertainty in key market and operational parameters. The results demonstrate that larger systems benefit from economies of scale, with lower per-unit costs. However, diminishing returns at larger scales highlight the need to balance technology design, processing scale, product valorisation, operational costs, and logistics. A sensitivity analysis reveals that gate fees, market prices for bio-based fertilisers and thermal energy fluctuations are critical variables influencing profitability. For Scale 3 (10,000 t/year) a reasonable balance between energy efficiency, cost-effectiveness, and logistical feasibility was observed. Still, the model showed susceptibility to market volatility, underlying the importance of adaptable strategies to mitigate financial risks and ensure system resilience in HTC systems. This research contributes to the circular economy literature, providing a transparent and adaptable framework for evaluating bio-based technologies under operational and market uncertainties. Future work should explore different reactor configurations, regional feedstock availability, and site-specific conditions to further validate and refine the system feasibility. © 2026 Elsevier Ltd

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Competitively-Won Funding Publications: SFI Fellowship

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Water use efficiency of Irish dairy processing

Yan M.-J.; Holden N.M.

Journal of Dairy Science, 2019

Dairy processing uses a significant amount of water for processing and cleaning. Withdrawing and distributing water and treating wastewater represent significant costs to the Irish dairy processing industry. Stringent discharge limits also add pressure for water use efficiency, particularly during peak production months. Improving water use efficiency is therefore critical for Irish dairy processing. We conducted a detailed analysis of water use efficiency in 4 Irish dairy processing plants. Using farm gate to processor gate (gate-to-gate) life cycle assessment, we assessed on-site water data quality and investigated gate-to-gate volumetric water use and eutrophication potential (EP) for 3 common dairy products. We also benchmarked the on-site water use and water balance, characterized wastewater nutrient load, analyzed the influencers of on-site water use, and identified scope for increased water use efficiency. We found that condensate from evaporation represented a significant input at the site level (0.51 to 1.14 L/L of fresh water purchased or extracted from nature). In terms of gate-to-gate volumetric water use, butter used 1,326 to 1,843 m³/t of solids, with electricity being the largest contributor, whereas milk powders used 3,006 to 3,754 m³/t of solids, with electricity and ingredients being the largest contributors. Eutrophication of butter was found to be 0.51 to 0.77 kg of PO₄ equivalents (eq)/t of solids, with transportation and nutrient emissions from wastewater treatment being the largest contributors. Eutrophication of milk powder was found to be 0.96 to 3.35 kg of PO₄ eq/t of solids, and contributions varied depending on powder specifications. Milk intake water use and various leakages were found to be hotspots that could be managed to reduce water use on site. Comprehensive metering is urgently needed to improve water use efficiency in light of the ongoing expansion of dairy production and hence processing in Ireland. Significant opportunities exist to optimize operator behavior, water reuse, and off-site transportation and energy. This study represents the first attempt to define water efficiency opportunities both at the site level and along the supply chain. Processors need to be aware of off-site contributors that significantly affect both volumetric water use and environmental impacts of processed dairy products. © 2019 American Dairy Science Association

Link to article: <https://doi.org/10.3168/jds.2019-16518>

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