PCP 1: Advances in Bioactive Peptides

Chairs: Hisham Ibrahim, Kagoshima University, Japan; Hitomi Kumagai, Nihon University, Japan; and Jianping Wu, University of Alberta, Canada

Hypocholesterolemic Pentapeptide Lactostatin (IIAEK) Activates Cholesterol Degradation via Hepatocyte Nuclear Factor 3α in HepG2 Cells
Satoshi Nagaoka*, *Gifu University, Japan*

Objectives: Novel cholesterol lowering milk peptide lactostatin (Ile-Ile-Ala-Glu-Lys: IIAEK) is the first identified hypocholesterolemic pentapeptide derived from bovine milk beta-lactoglobulin by our group (1). Lactostatin activates cholesterol 7α-hydroxylase (CYP7A1) concerning to the hepatic cholesterol degradation via L-type calcium-channel-related MAPK signaling pathway in HepG2 cells (2). However, the detailed molecular mechanism of the activation of cholesterol degradation is unclear. We tried to clarify the molecular basis of the activation of cholesterol degradation by lactostatin in HepG2 cells. Results & Discussion: Lactostatin significantly and specifically increased CYP7A1 mRNA levels in HepG2 cells. The protein level of CYP7A1 and its gene promoter activity determined by luciferase assay were increased by lactostatin. We have identified that the target promoter region of CYP7A1 gene mediated by lactostatin is the HNF3α responsive element. We found that HNF3α knockdown by siRNA inhibited the activation of CYP7A1 protein expression. Thus, HNF3α is an essential transcription factor for the activation of CYP7A1 gene transcription by lactostatin.

Bean Peptides Have High Binding Affinities for N-terminal Domain of Cholesterol Receptor Niemann-Pick C1 Like-1
Luis M. Real Hernandez*[1], and Elvira Gonzalez de Mejia[2], *[1]Ohio State University, USA; [2]University of Illinois, USA*

Hypercholesterolemia can be relieved by blocking the absorption of dietary cholesterol. Niemann-Pick C1 like-1 (NPC1L1) is an intestinal receptor that mediates cholesterol absorption. Pulse proteins, such as from beans, peas, and lentils, generate peptides due to gastrointestinal digestion, but the potential for pulse peptides to inhibit NPC1L1 has not been determined. In this study, in silico binding affinities and interactions were determined between the N-terminal domain of NPC1L1 and 14 pulse peptides (5≥ amino acids) derived from pepsin-pancreatin digestion. Peptides were docked to the N-terminal domain using computational docking program AutoDock Vina, and docking results were compared to those of ezetimibe, a prescribed NPC1L1 inhibitor. Four bean peptides (-7.2 to -7.0 kcal/mol) had higher binding affinities than ezetimibe (-6.6 kcal/mol) for the N-terminal domain of NPC1L1. Lentil and pea peptides did not have high binding affinities for the N-terminal domain. The common bean peptide Tyr-Ala-Ala-Ala-Thr (-7.2 kcal/mol) had the highest binding affinity. Ezetimibe and peptides with high binding affinities for the N-terminal domain are expected to interact at different locations of the N-terminal domain. The bean peptides Tyr-Ala-Ala-Ala-Thr, Glu-Arg-Ala-Phe, and Phe-Ala-Thr-Gly-Thr are all expected to have van der Waals interactions with SER130, PHE136, and LEU236 and a conventional hydrogen bond with GLU238 of NPC1L1. Due to their high affinity for the N-terminal domain of NPC1L1, bean peptides produced naturally in the digestive track have the potential to inhibit cholesterol receptor NPC1L1.

Absorption of Peptides into Rat Blood: Effect of Peptide-length and Aging of Rats on Absorption
Toshiro Matsui*, *Kyushu University, Japan*

It still remains unclear whether bioactive small

and oligo-peptides are absorbed into blood system as their intact form. Additionally, no reports on the effect of aging on their intact absorption have been found. In this study, we, attempted to clarify the effect of age on intact absorption of peptides into blood using young (8-wk) and aged (40-wk) spontaneously hypertensive rats (SHRs). For transport models in a single administration of peptide (10 mg/kg) to SHRs, designed peptides, Gly-sarcosine (Sar) as di-peptide and Gly-Sar-Sar-Sar as tetra-peptide, having protease resistance, were used, together with Trp-His and captopril as positive control. As a result, the tetra-peptide was detected in vivo in the plasma of tail vain for the first time, indicating that oligo-peptides with high protease resistance may be absorbable into blood system as intact form. The oral administration studies also provided useful information that aging of SHRs promoted the absorption of di-peptides used in this study, whereas the absorption of the tetra-peptide was not affected by aging. These findings would be explained by enhanced expression of PepT1 and no enhanced expression of tight junction-related proteins in intestinal membrane of aged SHRs, compared to young SHRs.

Enzymatic Processing and *in vivo* Actions of Anti-hypertensive Peptides Naoyuki Yamamoto*, *Asahi Group Holdings, Japan*

Hypertension is a major risk factor in cardiovascular disease, such as heart disease and strokes. In order to prevent disease incidence, pharmacological substances can be used to decrease high blood pressure to within the normal range. As functional food materials, many kinds of antihypertensive peptides originating from food proteins have been reported ever. Most of the reported antihypertensive peptides have inhibitory activities against angiotensin I-converting enzyme (ACE) that catalyzes release of the potent vasoconstrictor, angiotensin II from angiotensin I. Among reported bioactive peptides, antihypertensive peptides isolated from Lactobacillus helveticus fermented milk have been proven clinical effectiveness in our clinical studies. In the present study, Val-Pro-Pro (VPP) and Ile-Pro-Pro (IPP) generated from casein by fermentation with Lactobacillus helveticus are tried to prepare from casein by enzymatic treatment for industrial use of these active peptides. As the results, proline rich short peptides containing VPP and IPP were successfully developed. The casein hydrolysate showed a significant antihypertensive effects in many clinical studies. For the understanding of in vivo mode of actions, pharmacokinetical study was conducted after an oral administrations of both peptides to spontaneously hypertensive rats. From current our study, potential of these antihypertensive peptides will be discussed as the bioactive peptides.

Suppression of Melanoma Proliferation by an Amino Acid in Garlic Hitomi Kumagai*[1], Toshiki Ando[1], Tomoaki Yazaki[1], Hiroyuki Hara[2], and Makoto Akao[1] ,*[1]Dept. of Chemistry and Life Science, College of Bioresource Sciences, Nihon University, Japan; [2]Dept. of Dermatology, School of Medicine, Nihon University, Japan*

Garlic is known to have various physiological functions and the most effective food material to prevent cancer according to the report of Designer Foods Project performed at the initiative of the National Cancer Institute (NCI) in USA. The major active components are sulfides such as diallyl disulfide (DADS) and diallyl trisulfide (DATS) produced by C-S lyase from S-allyl-L-cysteine sulfoxide (ACSO). However, DADS and DATS are volatile lipophilic compounds having strong garlic odor, which limits their use in food. On the other hand, ACSO is a water-soluble amino acid and our previous findings have shown that orally-administered ACSO prevents platelet aggregation and hepatic injury. The present study was conducted to examine if ACSO prevents the

proliferation of melanoma, the most serious type of skin cancer. In *in-vivo* assay, melanoma cells were cultured in the medium containing ACSO, and the cell viability was evaluated by MTT assay. In *in-vivo* experiment, melanoma cells were injected to rats subcutaneously with oral administration of ACSO, and tumor growth was measured. In order to evaluate the effect of ACSO metabolites, the blood was collected from rats after oral administration of ACSO, and melanoma cell proliferation with the collected blood was measured by MTT assay. ACSO effectively suppressed melanoma proliferation both in vitro and in vivo.

Bitter Blockers: In Search of a Universal Bitter Taste Blocker Prashen Chelikani*, *University of Manitoba, Canada*

Humans can taste many compounds but are able to distinguish between five basic tastes, which are bitter, sweet, umami, salt and sour. In humans, bitter taste is sensed by a family of 25 G protein-coupled receptors, referred to as bitter taste receptors or T2Rs. The ligands that activate the 25 human T2Rs have diverse chemical structures and include peptides, natural alkaloids such as quinine, nicotine, and synthetic compounds. However, less than a dozen molecules are known to inhibit or block T2Rs with different efficacies. Recent studies from our group and others have shown that these blockers derived from animal, plant or synthetic in origin can block only a few but not all the 25 T2Rs. Pharmacological characterization led to the classification of one of these blockers, Nα,Nα-bis(carboxymethyl)-L-lysine (BCML) as an inverse agonist of T2R4. Interestingly, BCML does not block other T2Rs tested. I will discuss the conceptual advances on T2R structure-function studies, strategies and potential roadblocks in the quest to elucidate a universal bitter taste blocker.

Molecular Basis of Anti-inflammatory Action of Commercial Bromelain Hisham Ibrahim*, *Kagoshima University, Japan*

Bromelain is a mixture of proteolytic enzymes found naturally in the juice and stems of pineapples. According to some alternative medicine practitioners, bromelain is thought to have effects outside the digestive tract and is often marketed as a natural anti-inflammatory, for conditions such as arthritis, and nutraceutical preparations. While studies suggested positive correlation of the use of bromelain with reduction of symptom severity in osteoarthritis, the majority of the studies have methodological issues that make it difficult to draw definite conclusions. Due to a lack of research, bromelain has not been scientifically proven to be effective in any diseases and it has not been licensed by the FDA for the treatment of any other disorder. This study demonstrates that bromelain confers anti-inflammatory activity by suppression of pro-inflammatory cytokine, TNF-alpha, production from macrophage cells. It also exerts antimicrobial activity specific to Candida albicans. The results provide evidence, for the first time, that proteolytic activity does not contribute to the anti-inflammatory activity of bromelain and the action correlated to the generation of peptides through self-proteolysis. The finding may facilitate the rational design of promising complementary therapy and strengthen the knowledge needed for medical and nutraceutical practices.

Egg Yolk Antibody (IgY) Against Ornamental Carp's Pathogens and Its Prophylactic Effect Hajime Hatta*[1], Atsushi Sato[2], Kinjirou Morimoto[3], and Tomonori Somamoto[4], *[1]Kyoto Women's University, Japan; [2]Kyorin Co. Ltd., Japan; [3]Yasuda University, Japan; [4]Kyushu University, Japan*

Administration of specific IgY against fish pathogens would provide an alternative to antibiotic and chemotherapy treatment to prevent

fish from infectious diseases. IgY treatment is cost-effective and avoids slaughtering a stock of fish in order to stop the spread of diseases. We prepared IgY against ulcer disease of koi carp (Cyprinus carpio koi), which is a popular and economically valuable ornamental fish. Ulcer disease is caused by an infection of atypical Aeromonas salmonicid, resulting in skin ulceration and disfigurement. The primary sites of the infection are the skin and gills. Therefore, immersion of fish into water containing specific IgY was suggested as a preventive treatment. We succeeded in utilizing the anti-A. salmonicida IgY for prevention of the ulcer diseases of koi carp. Koi Herpes Virus (KHV) has known to cause 80-90% mortality in the common carp and Koi. Thus, it is important to detect and control KHV infections before its outbreak. We selected 3 candidate genes (ORF 25, 99, and 146) coding membrane glycoproteins, which are predicted to be responsible for KHV infection. Hens were immunized with three candidate recombinant glycoproteins and IgY was purified from each pooled egg yolk. Only the anti-ORF99 IgY showed significant neutralization activity against KHV infection onto KF-1 cells. Further experiments are required to confirm the anti-ORF99 IgY activity to prevent Koi from KHV infection for a prophylactic application of this IgY.

Scale-up Production and Product Development of Egg White Protein Hydrolysate with Angiotensin I Converting Enzyme Inhibitory Activity Jianping Wu*, and Qiyi Li, *University of Alberta, Canada*

Egg proteins are a well-known rich source of bioactive peptides with inhibitory activity against angiotensin I converting enzyme (ACE), a key enzyme responsible for the regulation of blood pressure. Peptides with ACE inhibitory activity could have potential application for prevention of high blood pressure, a chronic condition affecting about one third of Canadian adults. However, the lack of cost-effective methods of large-scale production and the presence of an unpleasant bitter taste limit the utilization of bioactive peptides and protein hydrolysates in functional food applications. The objectives of this study were to develop a scale-up method to prepare egg white protein hydrolysate with ACE inhibition activity and to investigate the applicability of egg white protein hydrolysate in two different food matrices. The optimal condition for preparing egg white protein hydrolysate was first determined by Taguchi's method and then applied for scale-up preparation. ACE inhibitory activity (expressed as IC50 value) and peptide yield of the egg white protein hydrolysates prepared in laboratory scale and large scale were 30 µg hydrolysate/mL and 77.5%, and 55 µg hydrolysate/mL and 53%, respectively. Egg white protein hydrolysate was incorporated into protein bars and protein beverages at up to 20% (w/w) and 2% (w/w), respectively. Protein beverages formulated with up to 1.5% (w/w) protein hydrolysate were found to be acceptable by participants, while all protein bar prototypes were not liked in general. Further research is needed to improve the consumer acceptability of protein hydrolysate.

PCP 2a: Advances in Protein Structure and Function

Chairs: Navam Hettiarachchy, University of Arkansas, USA; and Rotimi Aluko, University of Manitoba, Canada

Antioxidant Activities and Iron Binding Capacity of Protein Fractions from a High Protein Soybean Line Navam S. Hettiarachchy[1], Ali A. Bisly*[2], and Ronny Horax[2], *[1]University of Arkansas, USA; [2]University of Arkansas Fayetteville, USA*

Several new and specific lines and genetic traits of soybean have been produced recently. Soybean protein isolate (SPI) from a high protein seed line R95-1705, protein hydrolysate (SPH) obtained enzymatically by Alcalase hydrolysis, and its protein hydrolysate fractions (SPF) [$_{50}$ = 1.33 mg/mL); while SPH had the highest hydroxyl scavenging effect (25.0% and IC_{50} = 2.33 mg/mL). All protein fractions, SPF-1, SPF-2, and SPF-3, demonstrated remarkable activity against superoxide radicals at 1 mg/mL (55.1%, 53.3%, and 48.1% respectively). Tyrosinase inhibition activity was not observed in SPI, SPH, or SPFs. SPF-1 had the highest Fe(II)-binding ability which was 77.9% (IC_{50} = 0.658 mg/mL). These results demonstrate that protein hydrolysates of soybean seed line R95-1705, particularly SPF-1, has the potential for application in pharmaceutical and functional food products.

Interactions of Whey and Casein Proteins on Yoghurt Microstructure, Sensory, Tribology, and Rheological Behaviour Saara Laiho, Roderick Williams, Astrid Poelman, Ingrid Appelqvist, and Amy S. Logan*, *CSIRO, Australia*

A series of yoghurts were made from milks formulated for constant protein (5 %) and lactose (6 %) content and casein to whey protein ratios ranging from 80:20 (Control) to 50:50. Yoghurts were evaluated for their particle size, microstructure, rheology and tribology using instrumental techniques, and their sensory properties were evaluated by a trained sensory panel using a vocabulary of 19 attributes developed for visual appearance, texture in-spoon, in-mouth, and afterfeel. It was found that frictional properties and yield stress increased with an increasing phase volume of whey protein, resulting in a more heterogeneous microstructure observed through Confocal microscopy. Analysis by tribology suggests that the dominating factor causing friction in the Control yoghurt was the continuous homogenous protein network formed between whey proteins linked to casein micelles. However, the serum phase of the yoghurt had a greater influence on friction as the proportion of whey protein was increased, and the nature of the protein network changed to have larger voids between aggregates. Significant sensory differences were found for most attributes between the yoghurts containing the highest (50 %) and lowest (20 %) proportion of whey protein. Perceived creaminess and smoothness in-mouth decreased with whey protein content, yet no difference was detected in perceived thickness corresponding with yoghurt viscosity at 50 s-1. The ability to accurately predict the mouthfeel of stirred yoghurt using physical measurements will also be discussed.

The Use of Dairy and Plant Proteins in Foaming Applications and the Effect of Phosphates on Foam Capacity and Stability Jane Whittinghill and Sharon L. Book*,*ICL Food Specialties, USA*

Proteins are very complex polymers and widely used in food products for nutritional and functional purposes. Egg white proteins have been used for centuries in foaming applications and with advancement in separation techniques, dairy and plant proteins are being used to create a large range of foaming ingredients. The foam structure is greatly responsible for the texture and mouth-feel in foods such as angel food cake. Plant-based proteins such as pea and soy have found use in foaming applications but there is limited literature on their foaming behavior in complex food systems. A study was conducted to evaluate different dairy and plant-based proteins for foaming capacity and stability. The study looked at the effect of protein/sugar, protein/protein and protein/phosphate interactions on the foam integrity in a model angel food cake system. The ability of the proteins to foam was largely influenced by the water/protein ratio, acid, sugar, and phosphate.

Depending on the protein type, the addition of acid, sugar or phosphate to the model system either had a negative or positive effect on foam capacity and stability. SDS-PAGE confirmed the protein conformational changes that take place before and after baking. Understanding factors that influence foam structure is critical in formulating stable products using plant and dairy proteins.

Inhibition of ADAM17/TACE Enzymatic Activity by Rye Secalin-derived Peptides and Their Analogues Chinonye M. Udechukwu*[1], Apollinaire Tsopmo[2], Rong He[3], and Chibuike C. Udenigwe[4], *[1]Dalhousie University, Canada; [2]Carleton University, Canada; [3]Nanjing University of Finance and Economics, China; [4]University of Ottawa, Canada*

Inhibition of the enzymatic activity of "a disintegrin and metalloproteinase 17" (ADAM17) is an established therapeutic strategy to mitigating inflammatory diseases mediated by tumour necrosis factor-α. ADAM17 can be inhibited by chelation of the catalytic site zinc cofactor, which is required for substrate catalysis and structure stabilization. For the first time, this study investigated the ADAM17 inhibitory capacity of two tripeptides (CQV and QCA), derived in silico from rye secalin proteins, and their analogues (QCV and QVC). The peptides possessed a zinc-chelating capacity and dose-dependently inhibited ADAM17, with a maximum inhibition of 80%. Moreover, ADAM17 intrinsic fluorescence emission was quenched with increasing concentration of the peptides via static mechanism. Molecular docking revealed that the peptides extensively interacted with ADAM17 active site residues, and occupied mainly the S1 and S1' subsites. The peptides coordinated the zinc cofactor through their C-terminal carboxylate anions (QCV, QVC, and CQV) and carbonyl oxygen of the peptide bond (CQV). Extensive hydrogen bond interactions were observed, with the participation of the peptides' cysteine sulfhydryl group. Moreover, their hydrophobic contacts with the S1' hydrophobic pockets of ADAM17 suggest they are possibly selective inhibitors. The study findings indicate that food-derived peptides, particularly the secalin peptides, are promising candidates for therapeutic ADAM17 inhibition.

High-pressure Homogenization of Lentil Protein Isolates Significantly Influences Emulsion Stability and Emulsified Lipid Digestion Behavior Supratim Ghosh*, Maja Primozic, Akaysha Duchek, and Michael Nickerson, *University of Saskatchewan, Canada*

Lentil protein isolate (LPI) solutions (1 – 2 wt%) were pre-homogenized at 5,000 and 15,000 psi for 6 cycles before utilizing them to emulsify 5 wt% O/W nanoemulsions (20,000 psi, 6 cycles). Pre-homogenization significantly improved solubility and surface activity of LPI, while their surface hydrophobicity and interfacial storage moduli decreased. All nanoemulsions showed droplet flocculation and LPI aggregation, which was increased with protein concentration. The average droplet size of the nanoemulsions significantly decreased from ~250 nm for native LPI to less than 200 nm for pre-homogenized LPI solutions, while, no significant difference between 5,000 and 15,000 psi pre-homogenization observed. Although both the droplet size and instability indices of the nanoemulsions measured under accelerated gravitation increased after 28 days of storage, it was still much better than that from native LPI solutions. An *in vitro* static digestion model using simulated stomach and intestine conditions was used to test the digestibility of the emulsified lipid. A pH-stat titration was used to neutralize the amount of free fatty acids released during intestinal digestion, and it was observed that pre-homogenization of LPI significantly improved lipid digestibility. It was proposed that both the higher interfacial area of smaller droplets and the weaker interfacial moduli of pre-homogenized LPI-stabilized droplets were responsible for better proteolysis and removal of interfacial proteins by bile salts, leading to more accessibility of lipase towards the oil droplets. Pre-homogenization of LPI could be novel way to utilize pulse proteins in the formation and stabilization of nanoemulsions and improved digestibility under gastrointestinal conditions.

PCP 2b: Proteins for Delivery Functions

Chairs: Chibuike Udenigwe, University of Ottawa, Canada; and Lingyun Chen, University of Alberta, Canada

Food Protein Micro/Nano Particles for Controlled Nutraceutical Delivery in Functional Foods Lingyun Chen*, Jingqi Yang, and Cherry Yang, *University of Alberta, Canada*

The incorporation of nutraceutical compounds into food systems provides a simple way to improve public health and/or reduce the risk of disease. However, many of these compounds remain unavailable by oral administration, due to instability under the conditions encountered during food processing (e.g. high temperature, exposure to oxygen and light) or in the gastro-intestinal tract (e.g. low pH, digestive enzymes), and low permeability and/or solubility within the gut. Micro/nano encapsulation systems can be used to overcome these limitations. Although numerous synthetic polymer based delivery systems to maximize drug action and minimize side effects in the biomedical and pharmaceutical sectors have been successfully developed, these formulations cannot be used in food applications that require materials generally recognized as safe (GRAS). Food proteins as wall materials to encapsulate nutraceutical compounds are receiving growing scientific and industrial interest. Food proteins are GRAS materials with abundant natural sources and are widely used in formulated foods because they have high nutritional value and are degradable by digestive enzymes. Documented physico-chemical characteristics of food proteins indicate their excellent emulsifying and gel/film forming properties that offer unique potential in developing microcapsules for controlled release in food applications. This presentation focuses on the current knowledge and techniques for development of food protein-based micro/nano particles of desirable structures at precisely controlled sizes for target release of nutracetuical compounds at the site of absorption, as well as their potential applications in functional foods.

Chemistry Underlying the Preparation and Functionality of Protein-based Nanodelivery Systems Subin R. C. K. Rajendran*[1], Rickey Yada[2], and Chibuike C. Udenigwe[3], *[1]Dalhousie University, Canada; [2]University of British Columbia, Canada; [3]University of Ottawa, Canada*

Food proteins are one of the most widely available resources currently being utilized for the preparation of nanoparticles for delivery of bioactive compounds. Higher biocompatibility and amphipathic nature gives proteins, distinct advantages over other polymeric materials for the development of nanodelivery systems. In addition, proteins are unique in their molecular make-up and equipped with a variety of functional groups that can be selectively perturbed to modulate the encapsulation and release properties. A number of physical and chemical methods have been employed for preparing protein nanoformulations, each based on a different underlying protein chemistry. Physical methods (ultrasonication, high pressure homogenization, microfluidization) mostly result in conformational restructuring that stabilize multi-molecular nano/micro-structures whereas, chemical approaches generally ensure the formation of cross-linkages that reinforce the multi-molecular configuration in these structures or modify proteins (Maillard reaction conjugates) to enhance the stability or interactions. Proteins are also used in combination with other materials (polysaccharides such as chitosan, alginates) to improve functionality and provide a greater repertoire for us to prepare nanosystems with precise utility. The present study thus, focuses on describing the chemistry underlying protein nanosystems from the perspective of their preparation, functionality, stability and physiological behavior.

Simultaneous Encapsulation of Bioactive Nutrients based on O/W Emulsions Stabilized Whey Proteins Zheng Fang, Hao Cheng, Qi Fan, and Li Liang*, *Jiangnan University, China*

Functional foods containing bioactive nutrients offer benefits beyond basic nutrition and hence the possibility of delaying and preventing chronic diseases. Many bioactive nutrients degrade rapidly under food processing and storage. The key point to overcome

these limitations is to create a suitable edible carrier system for the encapsulation and protection of bioactive components. Milk proteins with high nutritional values have been widely used as carrier materials because of their ability to form emulsions and to interact with bioactive nutrients. Whey protein isolate stabilized oil-in-water emulsions have been widely studied, generally with hydrophobic nutrients dissolved in the inner oil phases. They were proved to be effective carrier systems for bioactive nutrients to improve their solubility and stability in aqueous solutions. Whey proteins containing beta-lactoglobulin and alfa-lactalbumin could reportedly bind vaious nutrients to form complexes. Therefore, bioactive nutrients (e.g. resveratrol) could possibly bind to protein membrane at the oil-water interface of emulsions. The data gathered suggest the potential to prepare the delivery system based on milk protein O/W emulsions for simultaneous encapsulation of multiple bioactive nutrients. These should be useful for the development of functional foods fortified with a range of bioactive nutrients and offering multiple health benefits.

Evaluation of Flaxseed Protein-polysaccharide Matrices for Encapsulation of Lipophilic Components

Xu-yan Dong*[1], Shanshan Du[2], Fang Wei[3], Xin Lv[2], Hong Chen[1], and Fenghong Huang[2], *[1]Chinese Academy of Agricultural Sciences, China; [2]Oil Crops Research Institute, Chinese Academy of Agricultural Sciences, China; [3]Oil Crops Research Institute, CAAS, China*

Flaxseed protein isolate (FPI) has lower solubility but better thermal stability than other oilseed proteins. Moreover, Zeta-potential, EAI and ESI of FPI stabilised emulsion was more than the WPI, SPI, Gel and SC stabilised emulsions. This study aimed to investigate the effects of mixing and conjugation processes on the emulsifying activity and other properties of FPI and different polysaccharide gum (pectin and flaxseed gum) in oil-in-water (O/W) emulsion and aqueous system. The effects of polysaccharide gum ratio and incubation time on conjugation were discussed. SDS-PAGE pattern and FTIR showed that hybrid conjugated polymers were formed. The zeta potential values and average particle size of FPI, FPI- polysaccharide gum mixtures and polymers were measured. During storage, the emulsion containing FPI-flaxseed gum conjugate was the most stable sample with the smallest droplet size among all prepared emulsions. This emulsion had the highest negative zeta-potential and stability among all samples. This study revealed that mixing and conjugation of FPI and polysaccharide gum led to improve the emulsifying activity of both polymers. These findings contribute to the development of FPI and polysaccharide gum complex as delivery vehicles for unstable and valuable nutrients such as omega-3 fatty acids.

PCP 3: Canola Proteins and Co-Products: Science and Utilization

Chairs: Curtis Rempel, Canola Council of Canada, Canada; Lisa Campbell, Canola Council of Canada, Canada; and Janitha Wanasundara, Agriculture and Agri-Food Canada, Canada

Production, Functional Properties, and Applications of Canola Protein Cruciferin Frank Pudel, Steffi Bäcker*, Jesus Palomino, and Ralf-Peter Tressel,*Pilot Pflanzenöltechnologie Magdeburg e.V., Germany*

Canola is one of the main sources of vegetable proteins worldwide. Today, the canola meal is used exclusively for animal feeding, although its nutritional value is also high for human nutrition. PPM developed a novel technology to produce pure rapeseed proteins. Cruciferin and Napin are the main storage proteins within Canola. Whereas, Cruciferin can be produced of a purity >90% with a simple extraction/precipitation method; Napin needs additional purification by chromatographic procedures. The high nutritional value, the available amounts, and the simple production enable the Cruciferin to be used in the food sector, whereas Napin could be used as food additive. In laboratory tests the functional properties, like solubility, foaming, and emulsification properties, were tested and compared to commercial products. Cruciferin is an alternative to the available plant protein sources. Potential applications in the food industry might be in the dairy, meat or dressings, and sauces sector.

White Flake Desolventization, Feedback from the Field Richard W. Ozer*, *Crown Iron Works, USA*

White flakes is a generic term describing hexane extracted oilseeds such as soy, canola, or sunflower that have been desolventized a low temperature to preserve protein value. Gentle desolventization of non-traditional oilseeds creates the possibility of a whole new range of products such as SPC, Canola & Sunflower Protein Isolate for the Food & Feed industry. This paper will describe the overall system and provide detail on the different methods of desolventizing the white flakes and with effect on Capital and Operating Costs. While the technology for White Flake Desolventization has been well known for over 30 years, there are relatively few plants exploiting this technology worldwide. Several startups for White Flake Desolventization plants occurred in 2015 or are scheduled for early 2016. We will relate some of the experience gained during these startups and potential impact on Capital & Operating Costs. Richard Ozer is Sales Manager for the Specialty Extraction Division of Crown Iron Works. He has been involved in the Pilot Plant Testing, Design and startup of Crown's Specialty Plants including SPC, algae, Krill for over 18 years.

Development of Protein-related Traits in Brassica Napus Danica L. Swaenepoel[1], Kenny So[1], Ashley Ammeter[1], Erin E. Higgins[2], Isobel I.A Parkin[2], Curt McCartney[2], Dwayne Hegedus[2], Janitha Wanasundara[2], Sally Vail[2], and Robert W. Duncan*[1], *[1]University of Manitoba, Canada; [2]Agriculture and Agri-Food Canada, Canada*

Developing canola with enhanced protein and nutritional qualities could revolutionize canola meal utilization and functionality. Canola meal has historically been a by-product and utilized only for animal feed, even though it has very similar available energy compared to soybean meal. This provides an immense opportunity to expand the utilization of canola in Canada. The Canadian canola industry contributes $19.3 billion to the economy annually; only about $525 million of this is composed from meal production and utilization. This value could grow several fold if high-quality protein products were developed for use in human food products. Protein-related traits may include protein, amino acid, cruciferin and napin contents. Cruciferin and napin are the two main seed storage proteins found in canola meal. Each of these have specific functional properties with cruciferin acting as an excellent emulsifying, gelling and binding agent. Napin provides strong solubility, the ability to form transparent solutions, foaming properties and excellent nutritional value. If the genes controlling seed storage protein type were known, genotypes high in cruciferin and/or high in napin could be developed and utilized in specialty food products. Collaborators and the Brassica Breeding Program at the University of Manitoba are working to determine the variation and the regions

controlling these protein-related traits. Genetic insight into these traits will provide the resources necessary to tailor canola genotypes for use in specialty, high-value protein products.

Production of Food-grade Canola Proteins by Membrane Based Processes Bih King Chen*, and Levente L. Diosady, *Dept. of Chemical Engineering, University of Toronto, Canada*

A patented process of protein isolation from edible oilseeds was developed by our research group. The process involves a series of well-designed unit operations: protein dissolution, chemical treatment, centrifugation, membrane separation (ultrafiltration and diafiltration), isoelectric precipitation, and drying. In the bench-top experiments, all of these individual steps were optimized to ensure the quality and yield of the final products. The process yields 3 products: a precipitated protein concentrate with 83% protein, a soluble protein isolate with 91% protein (both are on dry basis, N×6.25) and a meal residue with 26% protein suitable for animal feed. The isolates are light in colour, and bland in taste. They are essentially free of glucosinolates, and have very low phenolics and safe levels of lysinoalanine. The same process was scaled up to 50kg starting batches, in pilot facilities at Texas A&M, and also at Baltmere AG in Estonia where defatted European rapeseed (similar to canola seed, also very low both in erucic acid and glucosinolate) was used as starting material. The Estonian process was much more successful due to more suitable equipment and process schedule resulting in a precipitated protein concentrate with 84% protein and a soluble protein isolate with 91% protein.

Dehulling High Protein Canola Seed to Produce >58% Protein Meal Matthew A. Robinson*[1], Thomas G. Patterson[2], Patrick J. Nelson[2], and S. Patrick Adu-Peasah[3],*[1]Dow AgroSciences, USA; [2U]*

Canola meal trades at a discount to soybean meal in consideration of its lower protein content among other factors. To improve canola meal's protein content, decades long development of front end and back end dehulling process technology has proven technically feasible but has faced limited adoption due to uncertain economics and end-markets. Meanwhile in recent years, canola breeding programs at Dow AgroSciences have successfully obtained novel genotypes capable of producing seed with 45% crude protein on an oil-free, dry matter basis. In this work, front end dehulling processes are applied to high protein genotypes to yield meals with greater than 58% protein on an oil-free, dry matter basis, exceeding the protein content of low and high protein soybean meals. The process also provides for reduction of the ADF from 14.0% to 4.6%, on an oil-free, dry matter basis; a 67% reduction. Additionally back end dehulling was applied to meal produced from high protein canola seed followed by methanol extraction of the meats fraction, resulting in a canola meal with greater than 65% crude protein, enabling access to protein concentrate specifications. Such high protein canola meal and concentrate products can gain access to markets traditional dominated by soymeal, including swine and turkey.

Combined Effect of Pretreatment and Fungal Bioprocessing for Upgrading the Nutritional Value of Canola Meal Bishnu Karki[1], J.R. Croat[1], W.R. Gibbons[1], M.A. Berhow[2], and K. Muthukumarappan[3], [1]Dept. of Biology and Microbiology, South Dakota State University, USA; [2]Ag and Biosystems Engineering, South Dakota State University, USA; [3]USDA, ARS, NCAUR, USA

Presence of the anti-nutritional factors (ANFs) such as glucosinolates (GLS) is one of the limiting factors and can even be toxic at high ingestion levels. Furthermore, large amounts of GLS can reduce palatability for livestock and thus reduce intake and growth rates. Although with the advancement in breeding technology, canola with lower levels of GLS (< 30 µmol/g) and erucic acid (< 2%) have been developed, however, feed inclusion rates are still limited to ~30%. The goal of this research was to optimize a pretreatment and fungal conversion process to enhance the nutritional value of canola meal. Various combinations of physical/chemical pretreatments, fungal cultures, and incubation methods were investigated to metabolize GLS into cell mass, CO_2, or other non-toxic components. These treatments also served to hydrolyze canola meal fiber into

carbohydrates which were then metabolized by the fungi into single cell protein. Solid-state and submerged state fermentations of HE and CP meals were conducted using filamentous fungi. During the solid state fermentation, T. reesei achieved the greatest increase in protein content for HE and CP (~23%) canola meal and highest level of GLS reduction (<1 µM/g). Submerged incubation is most commonly used in industrial settings due to easier material handling and process control. Canola meals were either subjected directly to submerged incubation with the fungal strains, or were first saccharified with a cellulase enzyme cocktail and then incubated with the fungi. Aureobasidium pullulans (Y-2311-1), Fusarium venenatum and Trichoderma reesei resulted in the greatest improvements in protein levels in HE canola meal, at 21.0, 23.8, and 34.8%, respectively. These fungi reduced total GLS content to 2.7, 7.4, and 4.9 µM/g, respectively.In trials with CP canola meal, the same three fungi increased protein levels by 24.6, 35.2, and 37.3%, and final GLS levels to 6.5, 4.0, and 4.7 µM/g, respectively. Among the fungi evaluated, P. kudriavzevii was the only fungi able to significantly reduce ADF in both saccharified HE (6.5%) and CP (9.6%) canola meal. Fungal fermentation alone was not sufficient in degrading the fiber component of the meal, hence; HE and CP meals were subjected to the various pretreatments (extrusion, hot water cook, dilute acid, and dilute alkali) to determine if fibers could be made more susceptible to enzymatic hydrolysis. Following pretreatment, canola samples were subjected to submerged incubation with three fungal strains (A. pullulans Y-2311-1, F. venenatum NRRL-26139, and T. reesei NRRL-3653). The combination of extrusion pretreatment followed by incubation with T. reesei resulted in the greatest overall improvement to HE canola meal, increasing protein to 51.5%, while reducing NDF, GLS, and residual sugars to 18.6%, 17.2 µM/g, and 5% w/w, respectively. Extrusion pretreatment and incubation with F. venenatum performed the best with CP canola meal, resulting in 54.4% protein while reducing NDF, GLS, and residual sugars to 11.6%, 6.7 µM/g, and 3.8% w/w, respectively.

PCP 4a: Proteins from New and Minor Sources: Physicochemical, Nutritional, and Functional Properties

Chairs: Lamia L'Hocine, Agriculture and Agri-Food Canada, Canada; and Jane Whittinghill, ICL Food Specialties, USA

Protein Extraction and Characterization from Microalgae Halime Idakiev, Steffi Bäcker*, and Ralf-Peter Tressel, *Pilot Pflanzenöltechnologie Magdeburg e.V., Germany*

Today, microalgae are mainly used for production of high-priced products, such as dyes, antioxidants or unsaturated fatty acids, or in dried form as food supplements. However, as one of the major constituents of the microalgae biomass proteins (between 30 and 60% [w/w]), mainly enzymes, which have diverse functional characteristics, are not considered a quality product at the moment. Valuing proteins present in the microalgae will make the microalgae processing more profitable and is an important step in microalgae biorefinery. Therefore, the present study focuses on the recovery of the proteins in microalgae next to the recovery of the high price products. For this purpose, commercially available and economically interesting microalgae species are selected. In laboratory tests, the process for obtaining functional proteins as co-products of actual valuable substances (such as dyes) has been investigated. The nutritional and techno-functional properties of the obtained proteins have been studied. The recovered proteins could be utilized in food or technical applications. Thus, use of microalgae proteins as a high-value product increase the added value of the microalgae processing.

Functional and Proteomic Characterization of Protein Products from Defatted Cold Press Meals Özgenur Özdemir, Bilal Çakir, and Ibrahim Gülseren*, *İstanbul S. Zaim University, Turkey*

Due to their biological activities, cold press oils are increasingly utilized as functional foods or ingredients. Industrial extraction of cold press oils generates a significant amount of defatted meals with elevated protein content that requires valorisation. In this study, utilizing the defatted meals of a local producer; black cumin, pomegranate seed, grape seed, pumpkin seed, sesame and hazelnut protein isolates were generated using three different methods (alkali extraction-isoelectric precipitation, salt extraction, and micellar precipitation) and protein contents were quantified by Kjeldahl analysis. Molecular weight and/or pI distribution of soluble proteins were investigated using SDS-PAGE and 2D-Gel Electrophoresis. Based on in gel tryptic digestion, a wide variety of plant peptides were generated. The data obtained from MALDI-TOF/TOF-MS were analysed for protein and peptide identification. The protein contents of freeze-dried isolates were demonstrated to vary between 35-89%. Protein content of pumpkin seed and black cumin isolates were higher than the other samples. The functional characteristics of protein isolates including solubility, water and oil holding capacity, emulsification capacity and foam forming capacity were studied and compared to that of a commercial product (i.e., sodium caseinate). The functional properties of proteins were highly dependent on the source and medium conditions and in some cases, they were superior to that of caseinate. We have identified 24 novel proteins including enzymes and approximately 100 novel tryptic peptides that were not previously identified in protein databases. Further studies on the interfacial and/or bioactive properties of the protein isolates and their corresponding peptides are currently underway.

Extraction and Evaluation of Rice Bran Protein Concentrates Cecilia Abirached*[1], Carla Bonifacino[2], Eugenia Franco Fraguas[2], Darío Cabezas[3], Jorge Wagner[3], Luis Panizzolo[4], and Gonzalo Palazolo[3], *[1]PEDECIBA Química, Dept. de Ciencia y tecnología de los Alimentos, Universidad de la República, Uruguay; [2]Facultad de Química, Universidad de la República, Uruguay; [3]Laboratorio de Investigación en Funcionalidad y Tecnología de Alimentos, Dept. de Ciencia y Tecnología, Universidad Nacional de Quilmes, Argentina; [4]Dept. de Ciencia y Tecnología de los Alimentos, Universidad de la República, Uruguay*

The defatted rice bran (DRB), which is obtained after extracting oil from rice bran, contains highly digestible, hypoallergenic and high lysine-content proteins. However, it is used for animal feed, fuel or silica source. The aim of this work is to achieve a full DRB utilization, obtaining concentrates composed by proteins and polysaccharides and thus, with functional properties of both types of macromolecules. The DRB flour (DRB-F) was obtained by DRB sieving (0.355 mm). It was dispersed in distilled water and treated with thermophilic α-amylase and amyloglucosidase until the complete removal of starch. Fiber and proteins were precipitated by addition of ethanol (71% v/v, pH 4.5). The precipitate was dispersed at pH 8.0 in distilled water and lyophilized to give the concentrate (DRB-C). On DRB-F and DRB-C samples, moisture (drying, 105°C), ash (dry-ashing, 550°C), crude protein (Kjeldahl method, N×6.25), lipids (Soxhlet method), fiber (Protsky method) and starch plus sugars (by difference) were determined. These latter carbohydrates were efficiently removed from DRB-F (>98%), leading to two-times increase of protein and fiber contents for DRB-C sample (48.0 and 27.0% w/w, respectively). For the concentrate, protein solubility (bicinchoninic acid method) at pH 4.6 and 7.0 was near to 10.0%. This low value would be attributed to thermal stabilization made to the rice bran prior to oil extraction which leading to protein aggregation. Therefore, additional physical and enzymatic treatments must be performed on rice bran or the concentrates to improve the protein solubility and their related functional properties.

The Impact of Thermal Processing Methods on the Protein Quality of Pulses, as Determined by *in vivo* and *in vitro* Methodologies James D. House*, Adam Franczyk, and Matthew G. Nosworthy, *University of Manitoba, Canada*

Introduction: Pulses constitute the edible seeds derived from non-oilseed legume crops including beans, peas and lentils. Pulses typically contain 22-24% crude protein (as-is basis). However, the quality of the protein in pulses is generally limited by deficiencies in one or more indispensable amino acids or by factors (e.g. anti-nutritive agents) that impact the digestibility of the protein. The current approved method (US FDA) for measuring the quality of dietary protein is the Protein Digestibility-Corrected Amino Acid Score (PDCAAS), where digestibility is assessed in a rodent bioassay. The use of *in vitro* methods could present an alternative method for assessing digestibility and PDCAAS. Objectives: To determine the impact of thermal processing on the *in vivo* and *in vitro* PDCAAS values of protein contained in select pulse market classes. Methods: Composite samples of pulses (50 kg) were secured from Canadian suppliers, and included the major market classes of beans, peas, chickpeas and lentils. Sub-samples were processed via: a) Extrusion; b) Boiling; or c) Baking. Proximate analysis, *in vivo* (rodent bioassay) and *in vitro* (pH drop) protein digestibility, and full amino acid analyses were performed on each heat-treated pulse class. Results and Conclusions: Digestibility and PDCAAS values were highest for boiled or extruded samples, with baking (dry heat) yielding the lowest digestibility values (*in vivo* or *in vitro*), particularly for bean flours. The current data can be used to support protein content claims for cooked pulses.

Potato by-Products as a Source of Functional Protein Ingredients: Innovative Biocatalytic and Green Approaches Salwa Karboune*, *Dept. of Food Science and Agricultural Chemistry, Faculty of Agricultural and Environmental Sciences, McGill University, Canada*

Our recent research has been focusing on the development of innovative biocatalytic and “green” approaches towards the production of protein isolates, peptides and their corresponding glycated derivatives from potato (Solanum tuberosum) by-products. Potato is a vegetable whose high-quality proteins are underestimated. In addition to their high proportion in the essential amino acids, potato proteins possess angiotensin- converting enzyme-inhibitory potency, an ability to reduce plasma triglycerides associated with a reduced risk of atherosclerosis, and stimulate the release of the appetite regulating hormone CCK. In addition, potato proteins have long been considered not economically feasible due to the low protein content (27% dry matter) found in tuber. However, potatoes rank the second largest protein supplying crop grown per hectare following wheat. A non-destructive

biocatalytic approach for their extraction from potato pulp was developed in order to minimize functional losses and enhance quality. Furthermore, the conjugation of proteins with carbohydrates through the "green" Maillard reaction was investigated in order to improve their functional properties and expand their applications. The modulation of the glycation reaction rate to control the protein conjugation was achieved through the use of inhibitors. The presentation will focus on our recent "proof-of principle" results illustrating the feasibility and the efficiency of new biocatalytic and green processes for the production of innovative functional food ingredients, from potato by-products, whose potential health benefits are increasingly being recognized.

PCP 4b: New Protein Sources and Technology Advances for Protein Processing and Utilization

Chairs: Hui Wang, Iowa State University, USA; and Keshun Liu, USDA, ARS, USA

An Overview of the Advances in Protein Processing Technologies Jing Zhao*, *California State University, Los Angeles, USA*

Food proteins not only act as an important macronutrient in human diet but also contribute to the functionalities and stability of foods. Proteins have been widely used as an ingredient in food products and dietary supplements. The preparation process of the protein ingredients and foods may alter the structures and functionalities of the proteins. The objective of this presentation is to provide an overview of the advances in food protein processing technologies. Two topics will be covered in the presentation: 1) advances in extraction and purification techniques of proteins from various food sources; and 2) novel processing technologies of protein concentrates and isolates and their impacts on protein physicochemical and functional properties. Specifically, the protein extraction and isolation methods such as air classification (dry milling), solvent, alkaline, acid, salt, and enzyme-aided extractions, foam fractionation, membrane filtration, and ion exchange will be discussed. New processing technologies to be addressed include high-pressure processing, ultrasound, irradiation, pulsed electric field, supercritical fluid extrusion, and cold plasma treatment. Their impacts on protein quality and functionalities will be highlighted.

Wheat Gluten: Properties and Value-added Utilization Michael Tilley*[1], and Bruna Mattioni[2], *[1]USDA, ARS, CGAHR, USA; [2]Federal University of Santa Catarina, Brazil*

Wheat flour is unique in the ability to form a cohesive dough which retains gas which is essential to the formation of the variety of baked products consumed throughout the world. Flour storage proteins comprise about 8 – 14% of the mass of flour the remainder of which is predominantly starch. Wheat gluten proteins make up about 70% of the total flour protein and are readily separated from starch due to their hydrophobic nature, as such are a valuable co-product of the wheat starch wet milling process. A large polymer, wheat gluten is composed of gliadin and glutenin proteins each of which contribute to the unique properties of gluten polymer. These unique properties have been shown to be useful in food applications, and non-food bio-based products including adhesives, films and coatings, and biomedical materials. This presentation will review the properties of wheat gluten proteins, potential uses for bio-based products, how mechanical and heat processing affect its structure, and challenges that need to be addressed.

Edible Insects as a New Dietary Protein Source Changqi Liu*, *San Diego State University, USA*

In the context of increasing world population and limited water and farm land resources, improving the sustainability of food production is imperative to meet the growing demand for food. Using insects as an alternative food source has been heavily discussed in recent years. Insects have high feed conversion rate and low environmental footprint as compared to the conventional livestock, poultry, and fish. In addition, insects are good sources of nutrients, particularly proteins with well-balanced amino acid compositions. The high feed conversion rate and nutritional values of insects make them a feasible option to address the global issue of food security. This presentation will provide an overview of the recent advances in edible insect studies with an emphasis on their utilization as a new dietary protein source. Specifically, feed conversion, nutrient compositions, and the protein digestibility and functionalities of various edible insect species will be discussed. Processing technologies and potential challenges of utilizing edible insects as a food and protein source will also be addressed.

Leaf Protein Extraction from Oat Forage: Investigation into Factors Involved and Optimization Keshun Liu*[1], Qian Liu[2], and Mike Woolman[3], *[1]USDA, ARS, USA; [2]Northeast Agricultural University, China; [3]US Department of Agriculture, Agricultural Research*

Service, USA

Increasing cost and limiting availability of animal proteins have created a need to identify alternative protein sources for use as food and/or feed. Although considerable emphasis has been on conventional plant proteins, such as proteins from oilseeds, cereals and their by-products, leaf protein, an unconventional one, has also gained renewed interest on the prospect that its concentrate can be a valuable co-product from cellulosic biofuel production and can also mitigate the food versus fuel controversy. Earlier work focused mainly on mechanical extraction of proteins from fresh leaves, but solvent extraction from dried and ground biomass is the only alternative method in recovering proteins during cellulosic biofuel production. In this study, oat forage was grown and harvested locally, and then dried, ground into powder and solvent extracted for protein. Various factors, including sample particle size, solvent to solid sample ratio, alkaline solvent concentration, extraction temperature, centrifugation force, and enzyme treatment, were investigated. The objective was two-fold: to evaluate factors affecting protein extraction, and to determine maximum protein extractability under given conditions. Results indicate that all factors under investigation had significant effect on protein extraction from oat forage, with solvent to solid ratio and alkaline concentration as the most influential ones. The maximum protein extractability was around 78%, but the conditions that achieved this level required high solvent to solid ratio and high alkaline concentration. Thus, production of leaf protein concentrate from dried biomass could be economically unfeasible.

Pennycress Protein Isolate: Pilot Plant Production and Application in Films and Polymeric Composites Mila P. Hojilla-Evangelista*, Gordon W. Selling, Victoria L. Finkenstadt, and Roque L. Evangelista, *USDA, ARS, NCAUR, USA*

Pennycress (Thlaspi arvense L., Brassicaceae), historically a weed, is a crop whose seed oil is being developed as alternative feedstock for biodiesel production, with the protein in defatted meal (35% content) as the likely major co-product. We reported previously that pennycress protein isolates (PPI) have desirable nutritional and functional qualities. The current research scaled up the production of PPI using 5 kg of defatted press cake (PPC) obtained by prepressing and hexane extraction. Key steps were alkali solubilization (50 L aqueous NaOH, 1.5 h, 50°C), centrifugation, supernatant collection, acid-precipitation (pH 4.0), dissolution of precipitate in pH 7.0 water, ultrafiltration-diafiltration, and freeze-drying. One-half kg PPI (92.1% crude protein) was produced, which calculates to ca. 50% protein recovery. PPI still had 7 major polypeptides with MW 6-42 kDa, was least soluble (5% soluble proteins) at pH 5.5, and had maximum solubility of 70% at pH 10 (less soluble than lab-produced isolate). PPI films, produced using glycerol as plasticizer, were homogeneous and had good elongations (up to 170%) but low tensile strength (2-7 MPa). PPC or spent solids from protein extraction (PSS) were combined with polylactide (PLA) during melt extrusion to produce green polymer composites that could be used in nondurable/indoor plastics applications or short term products. PPC-PLA composites had reduced tensile strength and elongation as PPC amount increased, but had significantly improved ductility (strength and flexibility). PSS-PLA composites showed less flexibility at both 10% and 25% wt/wt fill. Therefore, the observed enhancement in ductility was conclusively attributed to the pennycress protein component.

PCP 5: General Protein and Co-Products

Chairs: Buddhi Lamsal, Iowa State University, USA; and Nandika Bandara, University of Alberta, Canada

A Review of Plant Proteins from Pseudocereals, Legumes, and Minor Crops and Their Use in Food Applications Jane Whittinghill*, *ICL Food Specialties, USA*

The global demand for plant-based proteins is growing at a rapid rate fueled by increased consumer demand for sustainable vegetarian and vegan offerings in the market today. Plant proteins from seeds, grain legumes, pseudocereals, and other minor crops are becoming great candidates to satisfy the growing demand for plant protein worldwide. A large number of crops containing medium to high protein content are still largely underutilized, many of which are able to fix nitrogen and be beneficial to the soil as well. This presentation will introduce the audience to the different types of proteins in the market today. The presenter will cover pseudocereals such as quinoa, amaranth, buckwheat, and teff and discuss the nutritional and functional attributes that make these plants important protein sources. Grain legumes and other minor protein sources important in tropical and subtropical countries have found their way into mainstream markets today. Knowledge of the type, structure, and functional behavior of the proteins is critical in any food system. These plant protein sources additionally contain other nutrients considered beneficial to the human diet such as minerals and "health preserving" components such as flavonoids. Understanding the functional and nutritional attributes of these plant proteins is critical to formulating products for a variety of market segments.

Influence of Bioprocessing Approaches on the Recovery and Physiochemical Properties of Salmon By-product Hydrolysates Subin R. C. K. Rajendran*[1], Zied Khiari[2], Chibuike C. Udenigwe[3], and Beth Mason[2], *[1]Dalhousie University, Canada; [2]Verschuren Centre for Sustainability in Energy and the Environment, Canada; [3]University of Ottawa, Canada*

Fish processing industry is one of the largest sources of food waste which is currently, largely underutilized, despite the high content of food-grade proteins. This study compares a novel cost-effective microbial method, involving lactic acid fermentation (LAF), with the conventional formic acid (FA) treatment and Flavourzyme (FL) enzymatic hydrolysis for valorizing fish processing waste. The different bioprocessing approaches were used to treat the ground salmon viscera (with high activity of endogenous enzymes). FL processing was carried out at the optimum conditions of Flavouzyme (37 C, pH - 7.0), whereas LAF and FA processing approaches relied upon production (LAF) or addition (FA) of organic acids to lower the pH and activate the inherent proteases in these tissues. Impact of processing parameters (enzymatic activity, microbial population, pH) on the yield of different fractions (fish oil, protein hydrolysate and residue material), physiochemical properties (surface hydrophobicity, free amino-nitrogen, sulfhydryl content) and antioxidant properties (ferric reducing capacity, metal chelation, glutathione (GSH) protection) was determined. Highest protein hydrolysate recovery (~57% by weight) was seen in LAF. Although highest protein content (~87%) in the recovered hydrolysate fraction and lowest amount of residual fraction (~4%) was observed for the FA treatment. In general, FL had better antioxidant activity with higher Fe chelation (73.45%) and ferric reducing capacity (27.7mM GSH equivalent). However, LAF yielded hydrolysates with the better GSH protection capacity. This study forms the basis for ongoing research towards economically viable formulations from salmon or fish by-products that benefits overall general health as well as the industry.

Structural and Functional Characterization of Enzyme-derived Antioxidant and Antihypertensive Flaxseed Protein Hydrolysates and Membrane-filtered Fractions Rotimi Aluko[1], and Ifeanyi D. Nwachukwu*[2], *[1]University of Manitoba, Canada; [2]Dept. of Human Nutritional Sciences, University of Manitoba, Canada*

This study was conducted to investigate the structure-function relationship underpinning the

antioxidant and antihypertensive properties of flaxseed protein hydrolysates (FPH), which were obtained following the enzymatic hydrolysis of flaxseed protein isolate (FPI). The FPH samples produced with various concentrations of the food-grade protease, thermoase-GL30 were tested for in vitro (renin and angiotensin-1 converting enzyme inhibition) antihypertensive properties. The in vivo blood pressure-lowering property of the two most active FPH samples and their corresponding membrane-separated fractions was subsequently evaluated in spontaneously hypertensive rats. Additionally, results from in vitro antioxidant tests indicated that although the fractions and hydrolysate samples were generally active in scavenging 2,2-diphenyl-1-picrylhydrazyl radical (DPPH.+), the low molecular weight fractions (< 0.05) reduced the ferric reducing antioxidant potential (FRAP) of all the hydrolysates. The metal chelation activity of the samples was robust in general with the 3-5 kDa fraction of the 2.5% FPH sample significantly (p

Greening-induced Oxidation of Sunflower Butter Cookies as a Function of Different Sweeteners and Storage Conditions Sihui Liang*, and Lilian M. Were, *Chapman University, USA*

The use of sunflower butter as a potential source of antioxidant and replacement for peanut and tree nut butters in baked foods is limited by the greening reaction under alkaline condition. This study focused on how different sweeteners (maple, agave, corn syrups, honey and xylitol), relative humidity (7, 42.8, and 81.8%RH) and oxygen conditions influence the greening, texture, reactants (free phenolic and protein), and antioxidant capacity in sunflower butter cookies by using image analyzer, HPLC method, Folin-Ciocalteu and ABTS assay, respectively. Cookies made with maple syrup had higher pH (about 9.0), moisture content, lower chlorogenic acid, total phenolic and antioxidant capacity and were greener (>80%) and softer compared to cookies made from agave syrup and honey. The largest and least decrease in protein content in cookies after baking was maple syrup and honey, respectively. In addition, cookies made with xylitol had highest tryptophan fluorescence intensity. Results from LC/MS showed that cookies stored at 81.8%RH condition and those made from maple syrup had higher chlorogenic acid-lysine adducts after 24 h, an indication that the moisture and water activity had positive correlation to greening and formation of chlorogenic-lysine adducts. In conclusion, greening induced by high moisture would be applicable where the greening color is desired and inhibition of greening could be done by reducing the moisture during processing and storage.

Adding Hydrolyzing Enzymes During Fermentation Step of Dry Grind Ethanol Process Affects the Process Favorably Lei Fang, Tong Wang, and Buddhi Lamsal*, *Iowa State University, USA*

Hydrolyzing enzymes, mostly carbohydrases, are used during early stages of dry-grind ethanol process to improve downstream ethanol or corn oil yield. Protease and or combination of hydrolases and proteases are usually not added during fermentation stage; our preliminary studies show that doing so improves ethanol and corn oil yields. However, the effects of such process modification on dried distillers grains with solubles (DDGS) are not clear. The physicochemical changes and of DDGS obtained from corn ethanol process with addition of acid-stable protease (Fermgen, F) and pectinase/cellulase (PC) during fermentation were evaluated. In-vitro digestibility of DDGS was also evaluated based pigs digestive model. DDGS from PC addition had higher protein content (26.81, db%) than DDGS from control (22.41, db%) and F (23.40, db%) treatments. Fermgen-treated DDGS had lower protein digestibility (63%), whereas, PC DDGS had higher protein digestibility (73%), compared to the control (69%). Both the additions of F and PC increased the non-detergent fibers(NDF) at 36.12% and 28.95, db%, respectively, compared with the control (23.39 db%). However, PC hydrolyzed cell wall components and made DDGS more digestible, even when PC DDGS had high fiber content and darker surface color than the control. The non-starch carbohydrate hydrolyzing enzyme might be a good choice for ethanol producers to produce more digestible DDGS, without negatively effecting ethanol volumes.

Palm Kernel Cake as a Valuable Source of Antihypertensive Proteolysate and Bioactive Peptides: An *in vitro* and *in vivo* Study Mohammad Zarei*[1], Nazamid Saari[1], and Azizah Abdul-Hamid[2], [1]*Faculty of Food Science and Technology, Universiti Putra Malaysia, Malaysia;* [2]*Universiti Putra Malaysia, Malaysia*

The aim of this study was to produce a valuable protein hydrolysate from palm kernel cake (PKC) as a by-product of palm oil industries for the development of natural antihypertensive proteolysate and bioactive peptides. Extracted PKC protein was hydrolyzed using different proteases (alcalase, chymotrypsin, papain, pepsin, trypsin, flavourzyme, and bromelain). Subsequently, angiotensin converting enzyme (ACE) inhibitory and DPPH• radical scavenging activities of each proteolysate were evaluated. Protein hydrolysates produced by papain after 30h hydrolysis exhibited the highest ACE inhibitory (70.9%) and DPPH• radical scavenging activity (73.5%) compared to the other hydrolysates. When normotensive rats induced with hypertension were fed the hydrolysate at a dose of 75 mg/kg bodyweight, their blood pressures stabilized considerably. In addition, fractionation of the most effective (potent) hydrolysate by reverse phase high performance liquid chromatography indicated a direct association between hydrophobicity and ACE inhibitory and radical scavenging activities of the hydrolysates. Isoelectric focusing tests also revealed that protein hydrolysates with basic and neutral isoelectric point (pI) have the highest radical scavenging activity, although few fractions in the acidic range also exhibited good antihypertensive and antioxidant potential. Nine peptide sequences were identified by Q-TOF mass spectrometry, and their respective ACE-inhibitory activities evaluated. The peptide sequences YLLLK, YGIKVGYAIP, and LPWRPATNVF showed ACE-inhibitory activities of 100%; however, the best IC50 values were observed for YGIKVGYAIP, GIFE and LPWRPATNVF at 1 µM, 3 µM and 20 µM, respectively.

PCP-P: Protein and Co-Products Poster Session

Chairs: Mila Hojilla-Evangelista, USDA, ARS, NCAUR, USA; and Navam Hettiarachchy, University of Arkansas, USA

1. **Egg-Derived Tri-Peptide IRW Promotes Differentiation of Mouse Osteoblastic Cell MC3T3-E1** Nan Shang*, and Jianping Wu, *University of Alberta, Canada*

Osteoporosis is a serious public health concern affecting more than 200 million people worldwide; it is also a major cause of morbidity and health expenditure in aging population. The tri-peptide IRW (Ile-Arg-Trp), previously identified from egg white protein ovotransferrin in our lab, has been investigated as a functional food ingredient in human health because of its anti-inflammatory, anti-oxidant and renin-angiotensin system (RAS) inhibition properties in vitro and in vivo. Recently, RAS has been proved that is directly involved in bone metabolism and plays important role in bone metabolic disorders. The overall objective of this study was to explore the in vitro potential benefits of egg-derived tri-peptide IRW in bone health management. Mouse osteoblastic cell MC3T3-E1 was incubated and treated with different concentrations of synthesized IRW (5 µM to 50 µM). The effect of IRW on cell differentiation was determined by western blotting and immunofluorescence. Our results showed that tri-peptide IRW could increase the expression of alkaline phosphatase (ALP) and type-I collagen (two major differentiation biomarkers), indicating that IRW could promote the differentiation of osteoblastic cell. Meanwhile, IRW increased osteoprotegerin (OPG) but decreased receptor activator of nuclear factor kappa-B ligand (RANKL) expression, implying the potential of IRW in bone resorption inhibition. In conclusion, this study showed the potential of egg-derived tri-peptide IRW in promoting bone formation but preventing bone resorption, which might open up a new application of egg-derived peptides against osteoporosis.

2. **Evaluation of Barley Protein based Nanoparticles for Vitamin B12 Delivery** Guangyu Liu* and Lingyun Chen, *University of Alberta, Canada*

Vitamin B12 is essential for human health. Severe vitamin B12 deficiency can potentially cause serious and irreversible damage, mainly to the brain and nervous system. Aged population is at a high risk of vitamin B12 deficiency, and more than 60 % of vitamin B12 deficiency is caused by food-cobalamin malabsorption syndrome due to gastrointestinal problems. Objective: This work aims to explore the feasibility of using barley protein as a carrier for vitamin B12 to improve its absorption. Method: Nanoparticles were prepared by high-pressure homogenizer. Their size, in vitro release profile, and safety were evaluated. Results: Nanoparticles with small size (250nm) and homogeneous distribution (PDI=0.24) were prepared. The release profiles were tested in the simulated gastro-intestinal tract. Release rate was low in the simulated gastric fluid, indicating nanoparticles were stable under gastric environment. In the simulated intestinal juice with pancreatin, the release of vitamin B12 was in controlled manner. Nanoparticles showed low cytotoxicity in Caco-2 cell model and significantly improved vitamin B12 cell uptake efficiency. In vivo tests in rat model indicated no significant change in organ mass and histology, blood biochemistry and intestinal function compared to the control group. Thus barley protein nanoparticles are relatively safe as a delivery system and have good potential to enhance vitamin B12 absorption.

3. **Surface Pressure Affects B-hordein Network Formation at the Air-water Interface in Relation to Gastric Digestibility** Jingqi Yang*, and Lingyun Chen, *University of Alberta, Canada*

Emulsions with controllable enzyme degradation profiles are desired in many applications, such in agriculture, food, cosmetic and pharmaceutics. Proteins are widely used as emulsion stabilizer due to their excellent biocompatibility and biodegradability. The digestion of emulsion was initiated from the degradation of the interfacial protein network. This study investigated how the conformation and orientation of the protein molecules at the interface

influence their digestibility. Langmuir-Blodgett B-hordein monolayer as a 2D model. B-hordein conformation and orientation under different surface pressures were determined by polarization modulation-infrared reflection absorption spectroscopy (PM-IRRAS). The interfacial network morphology was observed by atomic force microscopy (AFM). The results revealed that B-hordein interfacial network switched from an expanded liquid phase to a solid-like film with increasing compression pressure. Upon compression, the hydrophobic repetitive region of B-hordein tilted away from water phase. When compressed to 30 mN/m, a strong elastic network was formed at the interface, and it was resistant to a harsh gastric-like environment of low pH and pepsin. Based on these results, hordein was used to encapsulate hydrophobic bioactive compound β-carotene. Different from many hydrophilic globular proteins, hordein formed solid particles after high pressure homogenization and the hordein based β-carotene nanoparticles had less than 5% release in stomach. This work revealed how B-hordeins changed their conformation at the interface under pressure and how these changes influenced their digestibility. This knowledge was used to design hordein based β-carotene nanocapsules for controlled release in stomach.

4. **Influence of Structural Properties of Whey-derived Peptides on Zinc-chelating Capacity, and Simulated Gastric Stability/Bioaccessibility of Their Zinc Complexes** Chinonye M. Udechukwu*[1], Brianna Downey[1], and Chibuike C. Udenigwe[2],[1]*Dalhousie University, Canada;* [2]*University of Ottawa, Canada*

Zinc-chelating peptides from food proteins are relevant in human nutrition as dietary zinc delivery agents. Gastrointestinal stability of zinc-peptide complexes is essential for zinc delivery. As peptides' surface charge can impact the stability of their metal complexes, we evaluated the zinc-chelating capacity and stability of zinc complexes of whey protein hydrolysates produced with Everlase (WPH-Ever; ζ-potential, -39 mV) and papain (WPH-Pap; ζ-potential, -7 mV) during simulated digestion. WPH-Ever had lower amount of zinc-binding amino acid residues, but had higher zinc-chelating capacity than WPH-Pap, attributable to its highly anionic surface charge for electrostatic interaction with zinc. Zinc release during peptic digestion was lower for WPH-Ever, indicating higher gastric stability. However, over 50% of zinc remained bound in both complexes after digestion, and particularly for WPH-Ever. FTIR spectroscopy suggests the involvement of COO-, C-O of aspartate/glutamate, and C-OH of serine/threonine R-groups in the zinc-peptide complexation. These findings indicate that strong zinc chelation can concurrently promote gastric stability and impede intestinal zinc release, for peptides intended for use as dietary zinc carriers.

5. **Cholesterol-lowering Effect of Indigestible Proteins Isolated from Pulses** Hongyi Wu*[1], and Rotimi Aluko[2], [1]*Dept. of Human Nutritional Sciences, University of Manitoba, Canada;* [2]*University of Manitoba, Canada*

There is growing interest in the development of functional foods that can prevent or decelerate the progression of cardiovascular diseases (CVD). High blood cholesterol level is a well-known risk factor for CVD. A potentially safer and effective cholesterol-reducing method involves the use of protein materials that are resistant to gastrointestinal tract digestion. These resistant proteins have a highly hydrophobic character that enables cholesterol binding and elimination through the fecal route. In this study, soybean and pulses protein isolates were prepared and subjected to wet heat, dry heat, gelation and freeze-thaw treatments respectively before subjecting to consecutive digestion with pepsin followed by pancreatin. The digest was centrifuged and the residue that was resistant to proteolysis was freeze-dried. Cholesterol-binding ability, which was reflected by bile acid-binding capacity, was tested using 0.9 mL of 2 mM bile acid mixture added to 0.1 mL of 20 mg/mL of freeze-dried resistant protein suspension. The in vitro bile acid-binding activities of the resistant proteins were calculated as percent ratio of unbound bile subtracted from the total bile acid content. The mung bean resistant proteins (after dry heat) had highest bile acid binding capacity of 37.66%, which was similar to that bound by cholestyramine (standard compound). Additional results showed that the processing method used for protein isolate production had significant

effects on the yield and bile acid-binding capacity of the resistant proteins. Future work will determine the actual cholesterol-reducing ability of the resistant proteins using a rat model of hypercholesterolemia.

6. **Chemically Modified Canola Protein-nanomaterial Hybrid Wood Adhesive Shows Improved Adhesion and Water Resistance** Nandika Bandara*[1], and Jianping Wu[2], *[1]Dept. of Agricultural, Food and Nutritional Science, University of Alberta, Canada; [2]University of Alberta, Canada*

The potential of canola protein as an renewable polymer to develop biobased adhesives were explored in our recent studies. Similar to other proteins, canola protein also exhibit weak adhesion and water resistance. Exfoliating graphite oxide (GO) and nanocrystalline cellulose (NCC) at low addition levels (1% w/w NM/canola protein) proved to be effective in improving adhesion and water resistance. However, further improvements are essential in order to use canola adhesives in engineered wood products. The objective of this study is to develop a chemically modified canola protein-nanomaterial hybrid wood adhesives (CMCP-NM) with improved adhesion. Modification of canola protein with ammonium persulphate (1% w/w APS/canola protein) significantly improved ($p < 0.05$) dry strength of control from 6.38 ± 0.86 to 10.47 ± 1.35 MPa and wet strength from 1.98 ± 0.24 to 4.12 ± 0.64 MPa. APS induced protein crosslinking via Tyr-Tyr and Tyr-His interactions contributing to a covalently stabilized protein network might be the reason for improved adhesion. Exfoliation of NCC/GO in CMCP at 1% w/w (NM/canola protein) addition level further increased ($p < 0.05$) dry strength up to 12.50 ± 0.71 MPa and 11.82 ± 1.15 MPa where wet strength increased up to 4.79 ± 0.40 MPa and 4.99 ± 0.28 MPa for NCC and GO respectively. Synergistic effects of protein crosslinking, improved cohesive interactions, thermal stability and increased hydrophobic functional groups due to protein structural changes contributed to the improvement in CMCP-NM adhesive. CMCP-NM adhesive developed in this study shows a great potential for developing engineered wood products.

7. **Microbial Alternatives to Animal-derived Enzymes for Protein Processing** Kelly Gregory*, Caroline Best, Chris Penet, and Deborah Winetzky, *BIO-CAT, USA*

Trypsin and Pepsin are used as processing aids to hydrolyze proteins in order to alter their properties or to develop unique and value-added functionalities. Typically derived from bovine and porcine, Trypsin and Pepsin are subject to natural variations and supply fluctuations. Microbial-derived alternatives to both Trypsin and Pepsin have been identified and evaluated for their abilities to hydrolyze proteins. Six different proteins (casein, chicken, soy, tuna, wheat, and whey) were hydrolyzed at 40°C, pH 7 using Trypsin and microbial-derived Trypsin alternative. Four proteins (casein, chicken, soy and whey) were hydrolyzed at 40°C, pH 4 using Pepsin and microbial-derived Pepsin alternative. An N-acetyl-L-cysteine and o-phthaldialdehyde method for primary amino nitrogen (PAN) was used to measure the level of protein hydrolysis over time and create hydrolysis profiles for the different protein/enzyme combinations. An o-phthaldialdehyde method was used to measure degree of hydrolysis. PAN hydrolysis profiles for microbial Trypsin alternative equaled or exceeded Trypsin's on five of six different protein sources, but was deficient on casein. Degree of hydrolysis on wheat was about 60% for both enzymes. PAN hydrolysis profiles for microbial Pepsin alternative exceeded Pepsin on casein and equaled Pepsin on the other three protein sources. These studies demonstrate that microbial-derived proteases are suitable for use as an alternative to animal-derived proteases in protein processing. Microbial-derived enzymes typically have less natural variation and stable supply situations. In addition, the microbial-derived enzymes used in this study are non-GMO and suitable for use in vegetarian products.

8. **Effect of Food-Sourced Enzymes on Pulse Protein Digestion** Alberta Aryee*[1], and Joyce Boye[2], *[1]Delaware State University, USA; [2]Agric & Agri-Food Canada, Canada*

Recent studies have shown the effect of several mammalian digestive enzymes (MDE) used in various combinations either sequentially or simultaneously on

pulse protein digestion. In this current study, we investigated at the role of food-sourced enzymes (FSE) such as papain (from papaya), actinidin (from kiwi), and bromelain (from pineapple) in enhancing lentil protein digestion. In a two-stage model, variously processed lentil seeds (by dry-milling, cooking and isoelectric precipitation) were first pre-hydrolyzed with these three FSE and then with the MDE: trypsin, chymotrypsin and peptidase in the second stage. As observed from both the SDS-PAGE and SEC-HPLC results, pre-hydrolyzing lentil proteins with the FSE resulted in greater digestion of intact/high molecular mass protein in the order of: papain > bromelain > actinidin than after digestion with the MDE alone. The influence of the FSE also varied among the different processed lentil proteins. These FSE could act as digestive aid to improve digestibility and value of pulses as ingredients in food applications.

9. **Influence of Particle Size Distribution on Protein Digestibility and Functionality** Alberta Aryee*[1], and Joyce Boye[2], *[1]Delaware State University, USA; [2]Agric & Agri-Food Canada, Canada*

The nutritional, health-promoting and functional properties of pulses are well known. Very little is however known of the influence of particle size on digestion and functional properties. In this study, a Malvern laser diffraction particle size analyzer was used to investigate the particle size distribution (PSD) of dry-milled lentil flour (RLF), cooked lentil flour (CLF) and lentil protein isolate (LPI) and correlation with their digestibility and functionality. The dispersed flours and protein isolate showed wide ranges of sizes (<1 µm - 1 mm) and unimodal to trimodal distributions. LPI showed narrower distribution than CLF RLF. CLF had lower population of smaller and larger particles than the dry-milled RLF, and significantly larger particles and lower fractional population of smaller particles than the LPI. Digestibility of LPI was significantly higher than in both RLF and CLF and this can be ascribe to the greater surface area of smaller particles and greater exposure for substrate-enzyme interaction and digestion. PSD affected the water holding and fat absorption capacities, with both indices increasing when the particle size was reduced, resulting in higher surface area available for holding water and absorbing fat. The intact cells in the larger particles could explain the lower measured effects in the RLF.

10. **Food and Feed from Flies: Compositional Analysis** Alberta N.A. Aryee[1,2] and Beth Mason[2], *[1]Department of Human Ecology (Food Science and Biotechnology) Delaware State University, USA, [2]Verschuren Centre for Sustainability in Energy and the Environment, Cape Breton University, Canada*

Current methods of producing feed and food are not very sustainable coupled with the growing number of mouths to feed and the large volumes of food waste. One prospective solution for alternative food and feed and waste processing include the inclusion of edible insects in the mix, which wouldn't be entirely new since insects are already part of the diet in many countries and have been nourishing people around the world for several years. One of such insect is the black soldier fly (Hermetia illucens). Due to its high feed conversion efficiency, the valuable contributions of these insects are being revisited. The larvae from the hatched eggs naturally feed on and rapidly convert organic waste matter into protein and fat in a short cycle. Additionally these insects are not a pest or a vector for disease. In this study the effects of various diets on the proximate composition of black soldier fly larvae (BSFL) was evaluated. The diet included pre-consumer organic waste such as fruits and vegetables, potato skin as well as fermented food waste. Protein and fat content ranges between 39 - 40% and 7 - 30%, respectively. The most abundant fatty acid is lauric acid as well as high proportions of palmitic, myristic, linoleic and oleic acid. BSFL has the potential to meet nutritional, food, feed and food waste bioconversion challenges.