



Poster presentations

Biopolymer Assembly

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pBA-1

Structure of hyperbranched heteropolysaccharides of Acacia gum exudates: From spheroidal to prolate ellipsoidal conformation

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Acacia gum (GA), also called gum Arabic, is an edible dried gummy exudate obtained from the trunk and branches of *Acacia Senegal* and *Acacia Seyal* trees¹. It is harvested in arid and semi-arid regions all along a belt from Senegal to East Africa. Because of its functional properties, GA is unique among the natural gums and widely used in food and non-food industries as stabilizer, emulsifier, flavoring agent, thickener, or surface-finishing agent¹. GA macromolecules are hyperbranched, charged and amphiphilic heteropolysaccharides belonging to the arabinogalactan-protein (AGP) family. They are mainly composed of galactose, arabinose, rhamnose, glucuronic acid and a small fraction of proteins (1-3%). GA can be defined as a continuum of macromolecules differing by their biochemical (protein to sugar ratio, charge and hydrophobicity) and structural (hydrodynamic volume and molar mass) properties²⁻³. Despite their huge industrial applications, most of scientific studies focused on *Acacia senegal* gum. For instance, some differences in the chemical composition (sugar composition and protein content) between both *Acacia* gums were well highlighted. However, only the conformational properties of *Acacia senegal* macromolecules were well studied. Hence, this work focused on the characterization and the comparison of the structure and conformation of *Acacia senegal* and *Acacia seyal* macromolecules.

The analysis of glycosidic linkages evidenced a similar polysaccharidic backbone in *Acacia seyal* and *senegal* gums composed of 1,3-linked β -D-galactopyranosyl chains with side chains substitution in position 6. Despite this similarity, the degree of branching was found to be higher in *Acacia senegal* gum than in *A. seyal* gum (78.2% vs. 59.2%) with more branched galactopyranoses, shorter arabinosyl side branches, and more rhamnopyranoses in terminal position. The conformation of GA macromolecules was further characterized using SEC MALS experiments. *A. seyal* macromolecules appeared more structured and compact than *A. senegal* ones. For both *Acacia* gums, the anisotropy of macromolecules increased with the increase of the molecular weight; however, *A. senegal* macromolecules were the most anisotropic ones. The conformation of *A. seyal* macromolecules varied from spheres to oblate ellipsoids while *A. senegal* macromolecules varied from oblate ellipsoids to more anisotropic conformations, such as oblate and prolate ellipsoids.

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**pBA-2****Stabilization of oil-in-water emulsion by protein-polysaccharide colloidal particles**

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In recent years, there has been a significant progress in emulsion technology, which has resulted in innovative manufacturing of emulsion-based food products. In this paper, we demonstrate a new approach to stabilize oil-in-water (o/w) emulsions by food-grade particles. For this purpose, we synthesized the colloidal particles of whey protein isolate (WPI) and low-methoxy pectin (LMP) via electrostatic complexation, and demonstrated their use in the formation of stable oil-in-water emulsions. We found that WPI-LMP colloidal nanoparticles in different compositions had a z-average diameter in the range of 197-478 nm. These nanoparticles can produce stable, surfactant-free o/w emulsions with droplet sizes in the range of 0.76-1.56 μm at pH 5.0 and 4.5. According to the data of ζ -potential and analytical centrifugation, under conditions where the absence of LMP, the resulting emulsions are very unstable against creaming. In addition, pH and ratio of WPI-LMP play an important role in the stability of the emulsion, isoelectric's point of WPI induces a larger aggregation of protein and a higher concentration of WPI-LMP provides better stabilization to the emulsions against coalescence. Overall, this work will establish a new possibility of tailoring novel food emulsifiers as market oriented product development, in which the economical and potential aspects are also considered to create economically reasonable and reliable functional food ingredients.

Key words : food-grade particles, whey protein isolate, low-methoxy pectin, aggregation, oil-in-water emulsion.

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pBA-3

Structure and functionality of the supramolecular complexes of food biopolymers with PC liposomes mixed with docosahexaenoic fatty acid and stabilized by plant antioxidants

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By now it is well known that polyunsaturated fatty acids (PUFAs), in particular omega-6 and omega 3, are the most needed components in a daily diet for maintenance of a good health. However, there are the real challenges of using these PUFAs as food ingredients. First and foremost, because of their high susceptibility to an oxidative degradation during the food product preparation, transport, and storage, and, secondly, because of their low solubility in water and, consequently, in biological liquids in a body that can decrease their bioavailability. In order to tackle these difficulties we have tried to combine both the encapsulation ability and high solubility of such biopolymer amphiphilic molecules as both sodium caseinate (SC) and covalent conjugate ((SC) (Sigma Chemicals) + maltodextrin (SA2, AVEBE DE = 2), the weight ratio of SA2 to SC equals to 2 : 1)) with the protective abilities of a most effective natural plant antioxidants, namely, both ether oil of clove and oleoresins of ginger. As the main lipid object of our investigation we have chosen mutually complementary combination of the polyunsaturated lipids, providing the more healthy equimass omega-6/omega 3 PUFAs ratio: namely soy phospholipid (Lipoid S100: phosphatidylcholine (PC) (~ 59% of linoleic acid, ~ 3% of linolenic acid) with a pure docosahexaenoic (22:6) (DHA) fatty acid (Sigma Chemicals). The synergy in the protective action against the lipid oxidation of both the biopolymers and the plant antioxidants was found. In addition, the high solubility of the complex particles in an aqueous medium was revealed. The release of the polyunsaturated lipids in the simulated gastro-intestinal tract has been studied in vitro in accordance with «A standardised static in vitro digestion method suitable for food – an international consensus» (Minekus M., Alminger M., Alvi P., et. al., Food & Function (2014), 5, 1113–1124). Relying on the combined data of the static and dynamic multiangle laser light scattering, the particle electrophoresis, the atomic-force microscopy, and the electron spin resonance spectroscopy the structure-functionality relationships for the studied supramolecular complexes were analysed.

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**pBA-4****Microencapsulation of betalain microemulsion by complex coacervation in matrices of gelatin and gum Arabic**

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Betalains are nitrogen-containing natural pigments that provide bright coloration to fruits, flowers, and roots. Betalains are water-soluble and possess high antioxidant and free radical scavenging activities. Encapsulation of natural colors can be an interesting alternative for the replacement of artificial colorants for natural colorants in the food and pharmaceutical industry. The objective of this work was to encapsulate betalain-containing microemulsions by complex coacervation in matrices of gelatin and gum Arabic, and to evaluate betalain retention, particle size distribution, morphology, and colour.

Microcapsules were produced with wall material proportion of 1:1 gum Arabic:gelatin. The core material consisted of Tween 80:Span 80:propylene glycol; soybean oil:water (8:1:1:2:2, mass basis) microemulsions, in which the water phase was partially substituted for betalain extract (10, 20, 30, 40, and 50 %, mass basis). Betalain retention was determined by the ratio of total betalain content in the microcapsules after complex coacervation to the total betalain initially added to the prepared emulsion. After coacervation, the microcapsules were freeze-dried and analysed for particle size distribution, morphology, and colour evaluation.

Particle size distribution and average particle size were determined by laser diffraction using a dry powder dispersion unit. The morphology of the microcapsules was evaluated by scanning electron micrographs (SEM), and powder colour was analysed through CIEL*a*b* parameters. Betalain retention resulted in 33.2, 44.3, 71.7, 37.8 and 49.5 %, respectively, for samples with 10, 20, 30, 40, and 50 % of betalain extract in the microemulsion. All formulations had similar morphology with a typical structure of coacervate capsules. The particles presented closed structures, without cracks, indicating a complete coverage of the core materials. The size distribution curves exhibited bimodal behaviour. The first peak included diameters between 5 - 369 μm , while the second peak enclosed particle sizes between 709 - 2000 μm . The sample with 30 % betalain extract exhibited the highest dye retention (71.7 %), whereas the size distribution and the morphology of the samples did not show significant differences between samples containing different extract percentages. All the powders presented pink colour, which suggests that the pigment was embedded into the matrix.

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pBA-5

Characterisation of carboxymethyl cellulose, a cellulose gum from wheat bran

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Wheat bran is an abundant industrial by-product generated during the milling operation and represents an interesting biomass in food and feed industry. Cellulose from wheat bran was used as the raw material to produce carboxymethyl cellulose. Cellulose based materials are being widely used as they offer advantages like edibility, biocompatibility, barrier properties against moisture and oxygen, aesthetic appearance as well as being nontoxic, non-polluting and having low cost (Vasconez *et al.*, 2009). Cellulose was extracted from wheat bran at 85°C for 3 hours using 3% w/v of sodium hydroxide while hydrogen peroxide (H₂O₂) and tetraacetythylenediamine (TAED) at pH 11.6 were used as a bleaching agent. Carboxymethyl cellulose (CMC) is a linear, long-chain, water soluble, an anionic polysaccharide prepared from wheat bran cellulose in etherification process using sodium hydroxide and sodium chloroacetate, with isopropanol as a medium. Different concentrations of sodium hydroxide ranged from 5 to 30% w/v were used in this study. Characteristics of CMC such as CMC content, degree of substitution, molecular weight, solubility, moisture and viscosity were also investigated. CMC content decreased when the concentration of sodium hydroxide increased. The formation of sodium glycolate and sodium chloride as a by-product will affect the purity of CMC content and other characteristics of CMC. CMC extracted with 10% w/v sodium hydroxide, showed the highest value for degree of substitution with 0.84. Similarly, better viscosity and solubility compared to CMC extracted with other concentration of sodium hydroxide was obtained at 10% w/v.

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pBA-6

Cloudifiers in beverage manufacturing: Microparticles

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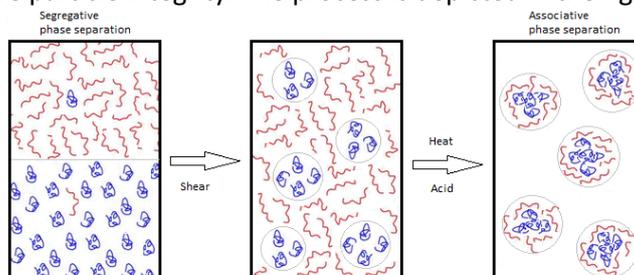
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In order to emulate the appearance of pure juice, artificial cloudifiers are added to beverages containing low amounts of fruit juice. Cloudifiers are colloidal dispersions rendering the beverage turbid in low amounts by scattering visible light. The scattering efficiency is influenced by the relationships between the wavelength of the light and the particles' size, shape, and concentration, as well as the relative refractive indices. Considering both colloidal stability and light scattering properties the optimal particle size is in the order of 0.5 μm [1].

Selection of ingredients and design of processing poses a challenge to beverage manufactures. Cloudifiers are often manufactured concentrated and subsequently added either to a beverage concentrate, which is then diluted prior to consumption, or to RTD beverages. The dispersed phase must be long-term stable in these different, continuous phases. In each step the viscosity, pH, ionic strength, density, and refractive index varies [1].

Common cloudifiers are oil-in-water emulsions produced from oils, weighting agents, emulsifiers, and stabilisers [2]. Alternatively, dispersions of solid microparticles (MP) such as proteins and polysaccharides (PPMP) can be applied. PPMPs were initially developed as fat-replacers; however often they possess optical properties making them suitable as cloudifiers. PPMP can be manufactured by sequential segregative and associative phase separation [3]. Heat is applied to denature the proteins and pH is lowered to ensure electrostatic complexation between the oppositely charged biopolymers [4], while shearing. A subsequent hardening step can preserve particle integrity. The process is depicted in the figure below.



The PPMP are sterically and electrostatically stabilised by the polysaccharide and may show good stability under acidic conditions and during pasteurisation [5]. The usage of MPs complies with consumer demand for cleaner labelling. Numerous patents for MPs from whey and casein have been filed. There has been a growing interest in MP from plant proteins, which have some advantages over animal proteins like generally being more sustainable, suited for vegan products, and less allergenic.

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pBA-7

Investigation of lipo & hydrocolloids in structuring edible oil

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Due to the adverse health effects that the consumption of saturated fats (SAFA) and *trans*-fats and its regular consumption has been linked to increase the incidence of cardiovascular diseases, high cholesterol levels, obesity, among other health problems. So most of the the lipid based food industries are looking for new functional substituents for products such as margarine, cream, shortening, pastries, etc. However, formulating food products without use of solid fats are quite challenging as they are responsible for providing the desired structure, texture, and mouth feel to the food products. Since solid fat has a wide range of technical roles, any fat reduction or replacement with an alternative material must ensure that these roles are still met. Therefore, we have recently developed a novel solid fat replacement, which was achieved by combing oil gels and hydrogels at a medium shear to obtain bigel systems. In this work, a new surfactant-free W/O emulsion fabricated by addition of plant wax into edible oils to acquire oil gels. Then followed by gelling the water phase with the help of a plant based polysaccharide, which together interacts very well in both oil and water systems. These combined hybrid systems are one of the promising drivers to limit saturated fats in lipid based food products. So these bigel systems could revolutionise the entire food manufacturers to reduce fat in baked foods such as cakes and cookies with out any compromise in sensory attributes. The main goal of this project was to modify the rheology of the aqueous and oil phase of a water-in-oil emulsion. Optical and confocal microscopy revealed the morphology of these emulsion, where starch molecules are embedded in the structured oil for which in turn results in softer texture of cakes. Therefore, our work demonstrated that a bigel system can be used to manufacture bakery items containing high levels of polyunsaturated fatty acids, which have been long recognized for its health benefits (e.g. prevention of cardiovascular diseases), potentially enabling 'reduced saturated fat' product claims to be achieved.

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**pBA-8****Velocity of rising air bubbles in aqueous beta-lactoglobulin solutions at different pH and salt concentrations**V. ULAGANATHAN¹, G. GOCHEV^{1,2}, C. GEHIN-DELVAL³, M.E. LESER³, R. MILLER¹¹ Max-Planck-Institute for Colloid and Interface Science, D-14476 Golm, Germany² Institute of Physical Chemistry, Bulgarian Academy of Sciences, 1113 Sofia, Bulgaria³ Nestlé Research Center, CH-1000 Lausanne 26, Switzerland

The local velocity profile (LVP) of a rising bubble can serve as a fingerprint for the dynamic behavior of the adsorption/desorption processes going on at a solution/air interface. Bubbles of air in pure water show a LVP with an range of acceleration and leveling off after a certain distance of the bubble movement. In solutions of surface active compounds at sufficiently high bulk concentrations, however, the bubble surface becomes very rigid due to the fast established adsorption layer and the observed terminal velocity is much lower as measured for a completely free and therefore mobile bubble surface. At intermediate concentrations a transition between the two extreme cases of the LVP are observed with a maximum velocity value the location of which is shifting to shorter distances with increasing bulk concentrations [1].

The LVP of bubbles in aqueous β -lactoglobulin (BLG) solutions proves to be extremely sensitive for the adsorption of proteins at very low bulk concentrations. In addition, it can show that the pH and ionic strength of the aqueous solution has a strong impact on the surface properties of BLG at the solution/air interface. This behavior is in line with experimental results at the surface of air bubbles at rest in BLG solutions [2].

It is observed that the time for establishing an immobile rigid surface layer at the rising bubble surface becomes shorter with increasing pH. A peculiar behavior is observed at the isoelectric point (IEP) where the LVPs show irregularities. Under dynamic conditions BLG has not its highest surface active at the IEP [3].

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pBA-9

Structure and viscosity of dense suspensions of fractal globular protein aggregates

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Solutions of the globular whey protein β -lactoglobulin (β -lg) were heated at different protein concentrations leading to the formation of polydisperse fractal aggregates with different sizes. The structure of the solutions was analyzed with light scattering as a function of the protein concentration. The osmotic compressibility and the dynamic correlation length decreased with increasing concentration and became independent of the aggregate size in dense suspensions. Results obtained for different aggregate sizes could be superimposed after normalizing the concentration with the overlap concentration. Dense suspensions of fractal protein aggregates are strongly interpenetrated and can be visualized as an ensemble of fractal 'blobs'. The viscosity of heated β -lg solutions increased extremely sharply above 80 g/L and diverged at 98 g/L, mainly due to the sharply increasing aggregate size. At fixed aggregate size, the viscosity increased initially exponentially with increasing concentration and then diverged. The increase was stronger when the aggregates were larger, but the dependence of the viscosity on the aggregate size was weaker than that of the osmotic compressibility and the dynamic correlation length. A different type of aggregate can be formed by heating β -lg in the presence of CaCl_2 . In this case homogeneous spherical microgels are formed. In comparison with fractal aggregates, the concentration dependence of the viscosity of solutions of microgels is much weaker, because they are much denser. The effect of mixing microgels and fractal aggregates has also been investigated

**pBA-10****Hybrid nanoliposome/electrosprayed encapsulation structures for the delivery of bioactive ingredients**

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Health benefits attributed to **functional foods** rely on the bioactivity and bioavailability of the active ingredients they contain, which may be lost during food processing, storage or digestion. A plausible option to overcome these limitations is to produce edible **microencapsulation** structures capable of protecting the integrity of the bioactive agents ¹.

Nanoliposomes, i.e. vesicles made of phospholipids surrounding an aqueous core, are promising delivery systems because of the similarity of their phospholipid bilayer to biological membranes. Moreover, they have been reported to improve the gastrointestinal absorption, bioactivity and water solubility of hydrophobic compounds. However, drug-loaded liposomes are often unstable, losing efficiency as the bioactive compounds leak out ².

Electrospraying is a versatile technology based on electrohydrodynamic processing which has been just recently applied for the microencapsulation of bioactive food ingredients within polymeric microstructures avoiding the need of employing high temperatures or toxic solvents. This technique produces a charged jet from a polymer solution/suspension flowing out from a capillary nozzle, by means of an electrical field, so that the solution is ejected towards a grounded collector. During the flight, the jet is elongated and finally breaks down into fine droplets, generating solid nano- and/or microparticles upon solvent evaporation ³.

The present contribution reports on the development of **novel hybrid microstructures** based on the use of nanoliposomes for primary encapsulation of bioactive ingredients, followed by a secondary encapsulation of these nanoliposomes within a protein matrix through electrospraying. This dual encapsulation strategy unifies the advantages of nanoliposomes as delivery vehicles with the preservation capability of electrosprayed protein particles ⁴, obtaining an easy-to-handle powdery ingredient.

More specifically, a whey protein concentrate has been used in this work for the microencapsulation of phosphatidylcholine liposomes containing curcumin as a model hydrophobic bioactive ingredient. The functionality and protection ability of the hybrid structures have been studied showing the potential of this type of hybrid systems for the delivery of bioactive molecules.

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pBA-11

Potential use of casein glycomacropeptide to design gelled emulsions as delivery of bioactive compounds

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Emulsion-based delivery systems is an advantageous encapsulating technique for incorporation of bioactive lipids or hydrophobic bioactive compounds in foods. As a delivery system, the emulsion-based structures as gelled could further protect the bioactive compounds against degradation. Casein glycomacropeptide (CMP), which is released from milk κ -casein during cheese making is a potential ingredient for specific dietary applications with several health benefits.

The aim of this work was to assess the potential of CMP to form and stabilize gelled emulsions alone or in combination with other emulsifiers. Sunflower oil in water emulsions were prepared using CMP and Tween 80, gelatin, lecithin and Arabic gum at concentrations between 2 and 8%. Pre-emulsions were prepared using ultraturrax and were then sonicated using an ultrasonic processor Vibracell Sonics model VCX 750, at natural pH (pH 6.5). In a second step the pH was decreased (pH 2-3) to promote the gelation of the emulsions, since CMP is able to gel at room temperature by effect of pH (Farías et al., 2010). By last, the gelled emulsions were reversed by increasing pH to pH 6.5 with NaOH 0.1 N.

The droplet size of the emulsions at pH 6.5 before and after the gelation, and the gel reversibility experiment was determined by static light scattering using a Mastersizer 2000. Also the stability of the emulsions was evaluated over time.

It was observed that single CMP emulsions were unstable, and the addition of other emulsifiers like Tween 80 (2 and 4%) and lecithin (2%) improved the stability (at least two months). The emulsions with CMP and Tween 80 showed the smaller droplet size (200 nm) and this value remained after the gel reversibility by pH (from 2-3 to 6.5), evidencing the great stability of the emulsions. These gelled emulsions have potential use as carrier of functional ingredients because besides using CMP which is a compound with bioactive properties, it is possible the use of bioactive oils and minerals that can be bound by CMP.

Farías, M. E., M. J. Martinez, and A. M. R. Pilosof. 2010. Casein glycomacropeptide pH-dependent self-assembly and cold gelation. *International Dairy Journal* 20:79-88.

pBA-12**Prediction of collapse time of polymer stabilized O/W emulsions**

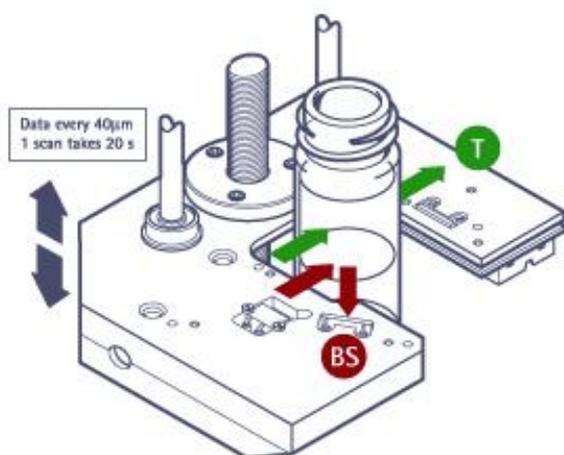
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Polymers are widely used in the industry as a tool to increase the stability. Depending on their concentration, they can act as depletion agents or gel agent. The stability of these systems is driven by the polymers and the structure of the network of droplets and can lead to collapse of the emulsions.

In this work, Multiple Light Scattering device is used to monitor the behaviour of w/o emulsions stabilized with polymers. The heart of the optical scanning analyser is a detection head, which moves up and down along a flat-bottomed cylindrical glass cell (see figure). The detection head is composed of a pulsed near infrared light source (wavelength = 880 nm) and two synchronous detectors. The transmission detector (at 180°) receives the light, which goes through the sample, while the backscattering detector (at 45°) receives the light scattered backward by the sample. The detection head scans the entire height of the sample, acquiring transmission and backscattering data every 40 µm.

We propose a description of the behaviour of o/w emulsions stabilized with different polysaccharides, we will show the advantages of using Multiple Light Scattering (MLS) to monitor their stability and propose a method to predict stability of these emulsions thanks to their size evolution in the first days after preparation.



Principle of MLS measurement

pBA-13**Improved stabilization of concentrated oil-in-water emulsions by complexing soy protein with κ -carrageenan**Iris TAVERNIER¹, Paul VAN DER MEEREN², Koen DEWETTINCK¹, Ashok R. PATEL¹¹ Lab of Food Tech. & Engg., Faculty of Bioscience Engg., Ghent University, 9000 Gent, Belgium² Particle and Interfacial Technology Group, Department of Applied Analytical and Physical Chemistry, Faculty of Bioscience Engineering, Ghent University, Coupure Links 653, B-9000 Gent, Belgium

Biopolymers such as hydrophilic proteins are commonly used stabilizers in water continuous emulsions. However, for the development of highly concentrated emulsions ($\phi_{oil} \geq 0.6$), the stabilization provided by only proteins often appears to be insufficient. The emulsion stabilization properties of proteins can markedly be enhanced by forming complexes with suitable polysaccharides to create colloidal particles that display surface activity. In this work, we report for the first time the use of soy protein isolate – κ -carrageenan complexes as stabilizing agents for concentrated oil-in-water emulsions prepared at $\phi_{oil} = 0.6$. The colloidal stability of concentrated emulsions stabilized with only soy protein isolate (SPI) was compared to emulsions stabilized by soy protein isolate – κ -carrageenan (SPI: κ -CG) complexes (particles) using a combination of techniques including advanced microscopy and light scattering experiments measuring the droplet size distribution. These measurements showed that a better long-term stability was achieved by using the SPI: κ -CG complexes compared to stabilizing the interfaces with only soy protein. Rheological studies revealed that the stability enhancement effect was due to interfacial accumulation of particles rather than an increase in the bulk viscosity. The interfacial accumulation of SPI: κ -CG particles was later confirmed from cryo-scanning electron microscopy images (Figure 1).

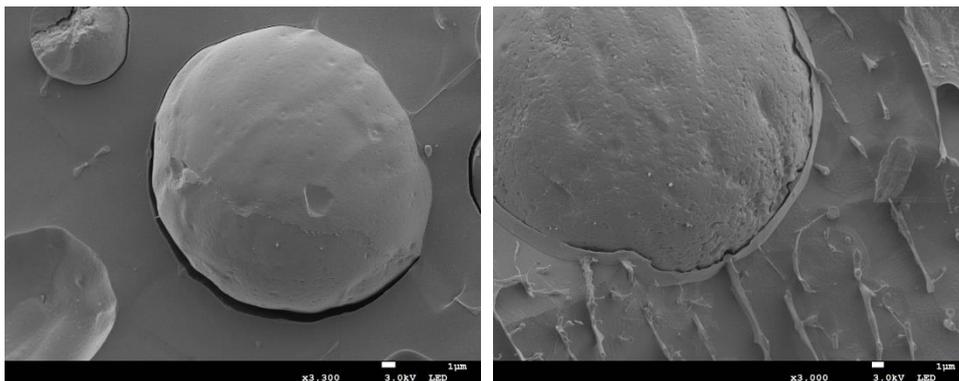


Figure 1: Cryo-SEM images of an emulsion droplet stabilized by soy protein isolate (left, smooth surface) and an emulsion droplet stabilized by SPI: κ -CG complexes (right, rough surface)

The potential of using SPI: κ -CG complexes as stabilizers for concentrated emulsions could open up new opportunities for the development of emulsifier-free (clean-labeled) food products since the majority of food products are based on concentrated emulsions.



pBA-14

Molecular Dynamics Simulation of Triglyceride Ordering at Surfactant Covered Triglyceride-Water Interfaces

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Molecular dynamics simulation (both all-atom and coarse-grained MD) has been used to simulate the interaction between glycerol monooleate (GMO) and tristearin (TS) at a tristearin-water interface and between polyglycerol polyricinoleate (PGPR) and tristearin at the TS-water interface. The simulations were carried out in an attempt to explain experimental observation made by Ghosh & Rousseau (2012) that suggest that adsorbed GMO promotes interfacial crystallization of triglyceride in water-in-oil emulsions whereas adsorbed PGPR does not.

Structure and ordering in the GMO, PGOR and TS layers are deduced through the calculation of the deuterium order parameter, lateral diffusion coefficient in the plane of the interface and the area occupied per head group for the surfactant molecules.

Formation of a GMO monolayer at the TS-water interface is evident in the GMO-TS-water systems. PGPR also adsorbs to the TS-water interface, but the structure of the interface differs from that of the GMO layer. Two mechanisms of interaction between TS acyl chains and the GMO layer are observed. Interpenetration of the TS acyl chains into the acyl brush of the GMO monolayer can occur, although this can be limited due to excluded volume constraints in the monolayer. This results in alignment and ordering of TS acyl chains along the axis of the GMO monolayer normal to the interface as they intercalate into the gaps in the GMO layer. In addition to this, if the GMO interface is densely packed, and there is insufficient space for the TS acyl chains to penetrate between the GMO acyl chains, they show a slight tendency to align parallel to the plane of the interface, i.e. at right angles to the alignment of the GMO acyl brush layer.

The PGPR forms a more disordered layer than GMO at the TS-water interface, and alignment of the TS acyl chains either normal or parallel to this.

The results are discussed in terms of a comparison to experimental results presented by Rousseau in a separate presentation.

Ghosh, S. & Rousseau, D. Triacylglycerol Interfacial Crystallization and Shear Structuring in Water-in-Oil Emulsions. *Cryst. Growth Des.*, 2012, 12 (10), 4944–4954.



pBA-15

Alternative crosslinking strategies for whey protein particles and the effect on the particle stability and functionality

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Colloidal protein particles are very promising vehicles to encapsulate, protect and release (bio-)active molecules in a controlled way in a complex food matrix. An elegant way to produce such particles is liquid antisolvent precipitation. This technique relies on the reduction of the quality of the solvent in which the proteins have been dissolved. The production of particles from water-soluble proteins typically takes place by introducing these proteins in an organic phase in which they are very poorly soluble. These particles, when harvested from this organic phase and reintroduced in water, however, tend to redissolve which hampers their application as encapsulation or delivery systems for food products. A popular, but non-food-grade way of increasing the particle stability against redissolution is by hardening the protein particles with glutaraldehyde.

The objective of this work was to explore the hardening potential of alternative aldehydes that are sometimes added to food systems as flavouring agents, such as vanillin, salicylaldehyde, syringaldehyde and cinnamaldehyde. Their effect on particle stability, functionality and *in vitro* digestibility was intensively studied.

All four alternative hardening agents proved to have some potential for stabilization of these particles against redissolution although cinnamaldehyde seemed to be the best performing, followed by syringaldehyde and vanillin. Salicylaldehyde was the least well-performing. When encapsulating anthocyanins in the protein particles followed by a hardening step, glutaraldehyde and - to a lesser extent - salicylaldehyde led to a significant decrease in anthocyanin color intensity. Furthermore, the hardening agents also affected the stability of particles under different conditions (pH, ionic strength and temperature-time treatments) and their *in vitro* digestibility.

The alternative hardening agents here used seemed very promising for hardening particles made with water-soluble proteins. This work will contribute to the popularization of protein particles as food-grade encapsulation systems for food applications.

pBA-16

Simple coacervation of Acacia gum solutions

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Acacia gum (GA) is a dried exudate obtained from the stems and branches of the trees of *Acacia senegal* and *Acacia seyal* (family Leguminosae) (FAO, 1999). GA macromolecules are hyperbranched acidic and amphiphilic polysaccharide-protein complexes. GA can be defined as a continuum of macromolecules that can be separated in at least three fractions (HIC-F1, HIC-F2 and HIC-F3) using Hydrophobic Interaction Chromatography (Renard et al., 2006). GA has a wide range of industrial applications such as flavor release and encapsulation by coacervation processes. The coacervation with GA concerned mainly the complex coacervation with the involvement of another partner

In this work, we investigated the simple coacervation of Acacia *senegal* gum and its two main HIC fractions (HIC-F1 and HIC-F2), by modifying its physicochemical conditions with the addition of ethanol. The cloud point of the solutions corresponding to the phase separation of water-ethanol-GA solutions was determined by measuring the absorbance at 600 nm (Figure 1 and Figure 2). For whole Acacia gum and its molecular HIC fractions, the amount of ethanol required to induce phaseseparation was reduced with the increase of Acacia gum concentration. Furthermore, less amount of ethanol was required to induce the phase separation in whole Acacia *senegal* gum than in its HIC fractions (HIC-F1 and HIC-F2). However, the cloud point of HIC-F2 showed to be closer to whole Acacia gum than HIC-F1, indication of the important role of the fractions on the cloud point. Phase separation occurred with the formation of Acacia gum coacervates (spherical particles). Upon centrifugation, the coacervate and supernatant phases were separated and further analyzed using FTIR and SEC MALS measurements. The coacervate phase was composed of GA macromolecules and water. Meanwhile, all the ethanol was recovered in the supernatant phase together with water and GA macromolecules. The molar mass distribution (M_w) of GA was modified around the cloud point with an enrichment of the coacervate phase in high M_w macromolecules rich in proteins. All these results evidenced subtle differences in the solubility properties between of Acacia Senegal and its HIC fractions.

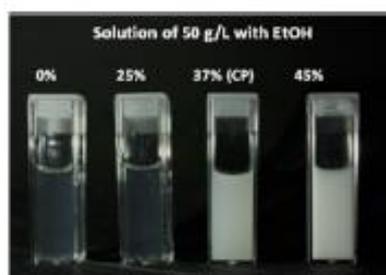


Figure 1. Simple coacervation of *A. Senegal*

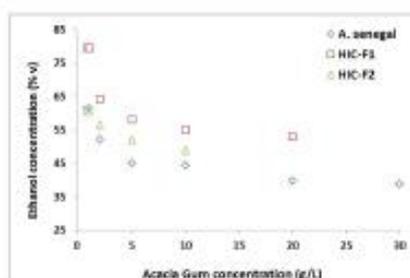


Figure 2. Phase diagram of the system *A. gum*/Ethanol/water

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pBA-17

Colloidal stability of polyphenols in young wine by Acacia gum: the major implication of arabinogalactan-proteins rich in proteins

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Acacia gum (GA), also called gum Arabic, is an edible dried gummy exudate obtained from the trunk and branches of *Acacia Senegal* and *Acacia Seyal* trees¹. GA macromolecules, belonging to the arabinogalactan-protein (AGP) family², are hyperbranched, charged and amphiphilic heteropolysaccharides composed especially of sugars (92-96%) and a small fraction of proteins (1-3%). GA can be defined as a continuum of macromolecules differing by their biochemical and structural properties³. Using hydrophobic interaction chromatography (HIC), three fractions could be isolated³⁻⁴. They were named HIC-F1 (85-94% of GA), HIC-F2 (6-18% of GA) and HIC-F3 (1-3% of GA), and classified in that order according to a growing hydrophobic index. Because of its functional properties, GA is widely used in food and non-food industries. In oenology, GA is used as additive to ensure the colloidal stability of young red wine precluding the precipitation of polyphenols. The efficiency of GA towards polyphenols instability is evaluated according to an "efficacy test" that consists to determine the quantity of GA required to prevent the flocculation of a colloidal iron hexacyanoferrate solution in hydro-alcoholic medium by calcium (International Oenological Codex).

In the present study, we investigated the stability mechanism of *Acacia senegal* gum towards the iron hexacyanoferrate – calcium flocculation and the efficiency of each HIC fraction in this mechanism. We evidenced that GA prevented the colloidal instability by the binding of calcium, the driving molecule of the flocculation, with the establishment of electrostatic interactions. For this GA, the critical concentration require to prevent the colloidal flocculation was 0.11 g.L⁻¹. Similar experiments performed with HIC fractions showed that the functional property of GA for this test was only based on HIC-F2 and HIC-F3 fractions with critical concentrations of 0.09 and 0.012 g.L⁻¹, respectively. Indeed, HIC-F1 fraction was devoid of functionality for this test. Further studies on a hydro-alcoholic matrix containing polyphenols also evidenced the functional properties of HIC-F2 and HIC-F3 macromolecules towards colloidal polyphenols instabilities. The functional properties of HIC-F2 and HIC-F3 towards colloidal instability could be in part ascribed to their biochemical properties as they are the most charged and hydrophobic fractions of GA.

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pBA-18

Extraction of Functional Proteins from *Chlorella* spp

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The microalgae *Chlorella* spp. is known to accumulate high protein levels in the dry mass, which depends on the cultivation parameters and the species. However, the whole cell shows a low functionality but the high protein content inside the cell might be a good source of functional proteins, which can be used in foods as e.g. emulsifier. To obtain the intracellular proteins from microalgae, the cells must be disrupted and by that the proteins are released from the different cell components.

We hypothesized that the method of disruption influences the extraction efficacy and the functionality of the obtained proteins. To study the cell disruption, food grade, and heterotrophic cultivated *Chlorella protothecoides* was chosen as a model organism with a protein content of 62 % in the dry matter. The cells were disrupted using different mechanisms including homogenization, enzymatic and chemical lysis, as well as thermal- and cryo-cracking. Depending on the method, efficacies of the cell disruption varied. Furthermore, the solubility and the macrostructure of the released proteins differed significantly. These results suggest that the disruption method has an influence not only on the cell disruption efficacy itself but as well on the functionality of the released proteins.



pBA-19

Gelatin dissolution is more affected by the structure than the water mobility

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Pig skins gelatin results of the collagen solubilization. Gelatin is used, amongst other applications, in the pharmaceutical industry to make hard capsules. Raw gelatin, on delivery, contains about 8-12% of water [1] and is a partially-crystalline polymer being composed of an amorphous phase (coil structure with primary chains) and a crystal phase [2]. The latter consists of partially reformed triple-helices of collagen. The shelf life of hard capsules is evaluated through gelatin USP dissolution test after aging under high temperature and high humidity conditions. After aging, the dissolution rate of gelatin is variable: either unchanged or reduced. The decrease of gelatin dissolution rate is partially due to intra- and intermolecular crosslinks formation in the amorphous phase which stabilize that structure [3]. Water in gelatin plays a role of plasticizer in the amorphous phase and is either free or bound to the polymer chains. The state of water in gelatin has been studied but no relation with aging or dissolution rate has been established. The aim of the present study was to understand how aging affects the gelatin structure and its water mobility, and in addition, to identify structural differences according to gelatin dissolution rate.

Thus, sixty pig skin gelatins with correct and non-correct dissolution rate were analyzed using Differential Scanning Calorimetry (DSC). Proton Nuclear Magnetic Resonance (¹H-NMR) spectroscopy was used on a subgroup of twelve gelatin films to measure the spin-spin relaxation time (T₂). For both methods, samples were taken before and after aging.

Aging decreased the amount and the stability to heating of crystals phase and increased the amorphous phase. The water mobility was increased which can be explained by the denaturation of crystals releasing the trapped water from the triple-helices [4]. After aging, the gelatins which failed the dissolution test showed a higher amount of amorphous phase but the water mobility was not modified. These results showed that the structure plays a major role in the gelatin dissolution rate. In order to support this major role of structural characteristics of gelatin, the conformation of the single chains and multiple helices will be assessed using circular dichroism.

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pBA-20

Spontaneous self-assembly of bovine collagen is modulated by age of animal and drying process

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Functionalities and bioactivity of collagen make this hydrocolloid of great interest for the food, cosmetic and pharmaceutical industry. Most important properties are those associated with the gelling behaviour, such as the texturizing, thickening and water binding capacity, and properties related to the surface behaviour, which include emulsion and foam formation and stabilisation, adhesion and cohesion, protective colloid function and film-forming capacity.

This study aims to evaluate the effects of bovine bone anatomy and age, and of the drying methodology on the self-assembly of collagen in the solid phase and on the aggregation in aqueous solution. Collagen (Figure a) was recovered from young (4 years) and old (7 years) cow's leg bones, namely femur and tibia (*Holstein* breed). Bones were milled and collagen leaching was then carried out with food-grade acetic acid 0.5 M during 5 days at 4 °C. Collagen solution was collected afterwards and the solvent was removed by two methodologies: spray-drying and freeze-drying (lyophilisation). The powders obtained were observed by scanning electron microscopy to evaluate the spontaneous self-assembly of collagen. An aliquot of the same powders were dissolved in 0.1 M acetic acid and at the concentration of collagen in solution after leaching (10 – 20 g/L depending on the bone)

Results showed that by spray-drying an assembly into micro capsules occurred and in a range 1 to ca. 20 µm (Figure b). Young bones collagen formed smallest particles, whatever the bone considered. On the contrary, lyophilisation gave rise to a spontaneous self-assembly into a tri-dimensional network (Figure c) whose uniformity and porosity varied according with bone anatomy and age. Collagen in solution showed an inverse behaviour depending on the drying methodology. A greater dimension was observed in the case of lyophilised collagen solutions unless for tibia old, which on the contrary generated the greatest aggregates in the case of spray-dried solutions.