Viscosity behavior of gluten free beta-glucan -PromOat® in oil-in-water emulsion

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MASTER THESIS

Lantmännen

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Lund, June 2024

Muhammad Kashif Inayat

Popular Science Abstract

Ever wonder what keeps your salad dressing creamy and plant-based beverages consistence? This thesis unlocks the secrets of stable emulsions!

Ever shaken a salad dressing and watched it separate? That's because oil and water naturally don't mix. But scientists have tricks up their sleeves to create stable emulsions, like the creamy delight in your dressing bottle. This research delves into the world of oat β-glucan, a natural ingredient with potential to keep these emulsions from breaking down.

The study explores these special oat fibers, focusing on a type called PromOat® that's gluten-free. It examines how GF PromOat® β-glucan behaves in oil-in-water mixtures. This Thesis investigates how well it works alongside a common emulsifier, soy lecithin, to prevent the oil from separating. It also focused on the best processing conditions to create the most stable emulsions possible.

This research could lead to the development of healthier and more natural food products with longer shelf lives. Imagine creamy dressings, dairy alternatives, or even spreads that stay fresh without needing artificial stabilizers! By harnessing the power of oat β-glucan, scientists are one step closer to creating delicious and functional foods that are good for you and the planet.

Abstract

This master thesis project was completed in partnership with Lantmännen, known for producing Gluten Free (GF) PromOat®—a concentrated oat β-glucan powder. The main aim of the thesis was to explore the properties of emulsions containing GF PromOat® oat β-glucan. It specifically focuses on the critical overlap concentration, viscosity, and stability before and after high-pressure homogenization (HPH) at 100, 300, and 700 bars.

The research was conducted in three primary phases: (1) identifying the critical overlap concentration of the hydrocolloid, (2) creating stable ingredient combinations, and (3) evaluating the impact of oat β-glucans on the stability of a reference oil-in-water emulsion using droplet-profiling techniques. Additionally, a comprehensive packaging evaluation of GF PromOat® was performed, which included an analysis of its packaging, labelling, and claims. This evaluation covered aspects such as containment, apportionment, and protection.

The critical overlap concentration was determined to be 0.0349 g/mL (3.49% m/m). Oat β-glucan solutions demonstrated shear-thinning behavior without forming gels. However, the oat β-glucan powder alone did not provide long-term stability. It makes the addition of an emulsifier necessary for kinetic stability.

Stability analysis using backscattering measurements revealed differences among the samples. It indicated that HPH conditions could causes a significantly impact in the emulsion stability. The results suggested that reducing oil droplet size during HPH enhances the stabilizing effect. Emulsions with oat β-glucans at or above the critical concentration remained stable for over 30 days when the HPH pressure was 700 bars. However, below this concentration threshold the depletion mechanism dominated. It overpowered the viscosity-induced stabilizing effect and resulted in instability. Future studies could investigate other food matrices, such as those with lower oil content.

Keywords: Gluten Free (GF) PromOat®, Oat β-glucan, Emulsions, Critical overlap concentration, High-pressure homogenization (HPH), Stability

Contents

1 Introduction

This chapter lays the groundwork, reviewing consumer trends and oat β-glucan properties. It then defines the research purpose and formulates key questions. Finally, it outlines the thesis structure for a clear roadmap.

1.1 Background

In recent years, the trend towards healthier and more sustainable food choices has gained significant momentum. Consumers are increasingly conscious of the ingredients in their food. They try to avoid extensive ingredient lists and those that seem unnatural. This heightened awareness has driven the food industry to reformulate many of their products. Many companies are launching new clean label alternatives to satisfy this growing demand. Producers face the challenge of eliminating or substituting certain ingredients without compromising on taste, texture, and shelf life.

Lantmännen, a Swedish cooperative, produces and sells Gluten Free (GF) PromOat®, a β-glucan derived from oats. These oat β-glucans are polysaccharides with both β (1-3) and β (1-4) linkages. The European Food Safety Authority (EFSA) has endorsed health claims related to β-glucans, including the maintenance of normal blood LDL-cholesterol levels and the reduction of post-meal glycemic responses. Besides their nutritional benefits, β-glucans are also used to enhance product quality and stability in various food items by improving their texture and appearance. This includes applications in sauces, salad dressings, cakes, bread, and ice creams.

1.2 Project Purpose and Research Questions

1.2.1 Purpose

The purpose of this thesis is to investigate the properties of emulsions incorporating GF PromOat® oat β-glucan, developed by Lantmännen. The goal of this thesis is to evaluate the functionality of oat β-glucans in GF PromOat® as an additive in food emulsions. Specifically, this project investigates the stabilizing capability of GF PromOat® in high-oil formulations, particularly emulsions containing 25% rapeseed oil. The research aims to determine the optimal range of GF PromOat® and soy lecithin, and to identify appropriate processing conditions for achieving stable emulsions. Additionally, the project includes a comprehensive evaluation of GF PromOat® packaging to ensure effective containment, apportionment, and protection against external factors that could affect product quality and integrity.

1.2.2 Research Questions

This thesis addresses several key research questions to thoroughly investigate the functionality and impact of oat β-glucans in GF PromOat® as an additive in food emulsions:

1. What is the critical overlap concentration (c) of Gluten Free PromOat®?
2. What is the optimum ingredient combination to make stable emulsions?
3. What is the impact of β-glucans and processing conditions on the stability of a reference oil-in-water emulsion?
4. How effective is the packaging, labelling, and claims evaluation of Gluten Free PromOat®?

By addressing these questions, the thesis aims to provide comprehensive insights into the application and benefits of GF PromOat® in food emulsions, contributing to the development of healthier and more stable food products.

1.3 Thesis Structure

The thesis begins with an Introduction, which provides the background context for the research, outlines the project's purpose, and formulates the research questions to be addressed. This section also delineates the overall structure of the thesis. Following the Introduction is the Literature Review, which comprehensively explores various aspects of oat β-glucan, including its physicochemical properties, functionality, health effects, and its role in emulsions and instability mechanisms, as well as techniques for stabilizing emulsions. The Methodology chapter details the materials used and the methods employed in the research, encompassing sample preparation, evaluation, and specific techniques like viscosity measurements and stability testing.

Results and Discussion form the core of the thesis, where findings from the research are presented and analyzed. This section covers the calculation of critical overlap concentration for GF PromOat®, viscosity-shear profile measurements, assessment of different ingredient roles in emulsions, and the evaluation of emulsion stability, including variations due to high-pressure homogenization (HPH) conditions. Finally, the thesis concludes with a chapter on Conclusions, Limitations, and Recommendations, summarizing the key findings, discussing any constraints encountered during the research, and offering suggestions for future studies. AI was utilized to correct grammatical mistakes in the writing process of the thesis.

2 Literature Review

This chapter reviews the literature on oat β-glucan, detailing its physicochemical properties, functionality, health benefits, and the advantages of Gluten Free PromOat®. It examines the mechanisms of emulsion instability and methods for stabilization, including homogenization and the use of soy lecithin as an emulsifier, with a focus on the stabilizing properties of GF PromOat®-oat β-glucan.

2.1 Oat β-glucan

β-glucan from oats is a prominent non-starch polysaccharide. It is made up of interconnected β-d-glucopyranosyl units bonded through β-(1 → 4) and β-(1 → 3) linkages. Oat β-glucan is sparking considerable interest as a potential functional food ingredient (Sun et al., 2020). It is derived from oat kernels and this viscous polysaccharide is predominantly concentrated in the endosperm cell walls. It is particularly adjacent to the aleurone layer and within the sub-aleurone layer (Daou & Zhang, 2012). Oats typically contain between 4.5% to 5.5% of β-glucan (Wood et al., 2011).

2.1.1 Physicochemical properties

Oat β-glucans are chains of sugar units linked together in a particular pattern, consisting of both (1→3) and (1→4) connections. These linkages depending upon the arrangement of connections and the size variance of the chains define the molecular characteristics of oat β-glucans. The distribution of (1→3) and (1→4) linkages within oat β-glucans is not haphazard; rather, it exhibits a semi-random pattern which reveals a dual organization level (Burton et al., 2010).

Through enzymatic analysis (figure 1), it has been found that a significant portion (roughly 90%) of oat β-glucans comprises cellotriosyl and cellotetraosyl units connected by (1→3) bonds. These units resemble cellulose and consists of three or four sugar units linked together in a (1→4) arrangement. The remaining section of the β-glucan consists of cellulose-like structures with a Degree of Polymerization (DP) ranging from five to 13–16 (Wang et al., 2003). Consequently, the structure of oat β-glucans is characterized by nonrandom sequences of sugar units.

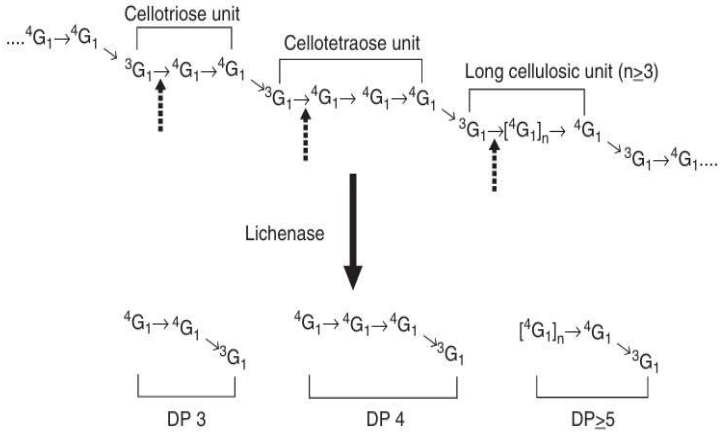

Figure 1; Cereal-glucan general structure and lichenase-mediated debranching of them; dotted arrows point to the lichenase hydrolysis sites in the polysaccharide chain (Lazaridou and Biliaderis, 2007).

2.1.2 Functionality

The food and nutritional supplement sectors are becoming more and more interested in using β-glucan as a component in product formulations (Vasanthan & Temelli, 2008). β-glucan can change physiological reactions like postprandial blood glucose response (PBGR) when added to cereal products (Henrion et al., 2019). It exhibits several advantageous functional characteristics, including thickening, stabilizing, emulsifying, and gelling capabilities (Ahmad, et al 2012). Thus, its application might be expanded to add thickness and stability to variety of culinary items. These include salad dressings, gravies, ice cream, and beverages. It is also used in infant foods, soups, sauces, and low-fat sausages. Additionally, it can be applied to dairy products as well (Lazaridou & Biliaderis, 2007).

Oat β-glucan viscosity-enhancing properties can be influenced by factors such as concentration and molecular weight, which could affect rheological behavior significantly (Wood et al., 2011). Furthermore, molecular weight influences rheological properties, as seen in non-Newtonian fluid behavior within the concentration range of 0.2–3% (Shen and Yao, 2005). At concentrations above 2 g/L, β-glucan exhibits pseudoplastic behavior that means decreasing viscosity with increasing shear rate (Dongowski et al., 2005). Moreover, its water-holding capacity and gelling ability contribute to its multifunctionality (Hu et al., 2009). Additionally, the rheological properties of oat β-glucan could be influenced by factors like concentration, temperature, and pH (Shen and Yao, 2005). Within a narrow concentration range, 1-2%, β-glucan transitions from homogeneous to heterogeneous behavior, highlighting its complex nature (Autio et al., 1987).

In different food formulations oat β-glucan perform different roles. It improves the moisture retention in baked goods to prolong shelf life and enhance texture (Beer et al., 1996). Its shear-thinning characteristics facilitate spread ability in low-fat products, such as cream cheese, without compromising stability (Wang et al., 2008). Additionally, β-glucan's odorless nature ensures it does not affect the flavor of food products, making it applicable across a wide range of formulations (Beer et al., 1996).

2.1.3 Why gluten free PromOat®

GF PromOat® is produced in two main ways. Dry milling grinds dehulled oat groats into flour while wet milling uses enzymes to break down oat bran into a liquid beta-glucan extract. Sterilization and decanter are used to make Oat β-glucan slurry. The slurry is then processed through enzymatic treatment to make GF PromOat® as described in figure 2. The introduction of PromOat® Gluten-free Organic marks a pivotal addition to Lantmännen's esteemed PromOat product line (Lantmännen Biorefineries, 2024).

GF PromOat® has been developed to accommodate a diverse range of culinary applications, making it an ideal ingredient for incorporation into foods, beverages, and dietary supplements that possess cholesterol-lowering properties. Additionally, it functions as a clean-label texturizer as well. GF PromOat® stands out as a leading gluten substitute, particularly benefiting individuals with gluten intolerance. By replicating the viscoelastic properties of gluten in gluten-free formulations, Gf PromOat® eliminates the necessity for other hydrocolloids (Lantmännen Biorefineries, 2024).

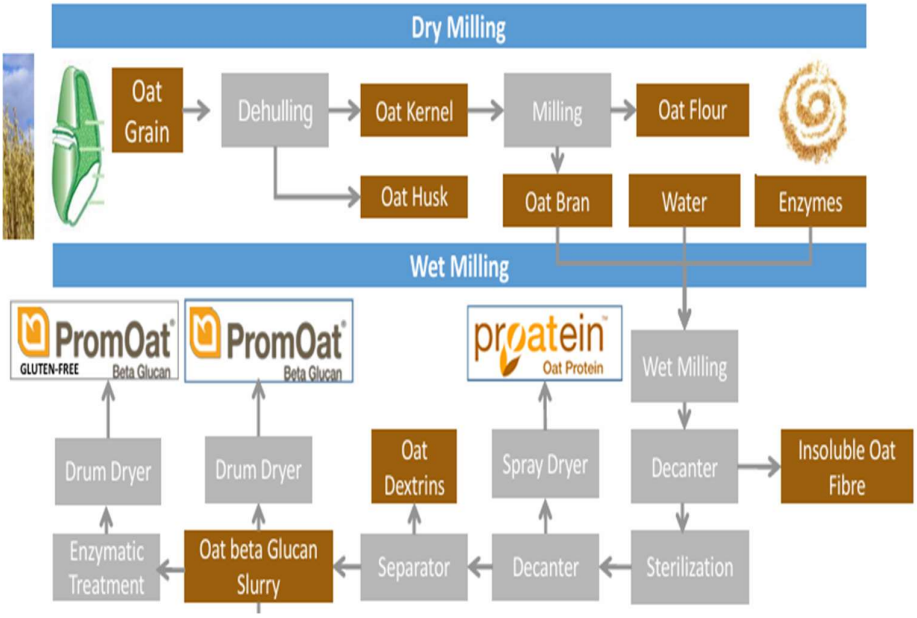

Figure 2; Flowchart showing the extraction of GF PromOat® (Lantmännen)

2.1.4 Effects on health

Numerous studies in scientific literature have highlighted the myriad health benefits of β-glucan. Since the 1970s, research has delved into the health effects of oat β-glucans, with significant findings (Ahmad et al., 2012). These advantages mostly result from the dietary fiber content of β-glucan. Dietary fiber is resistant to digestion in the small intestine but is partially fermented by microflora in the large intestine. Soluble dietary fiber (SDF) and insoluble dietary fiber (IDF) are the two functional categories of dietary fiber. The oats' extracted β-glucan contains both types of dietary fibers (Guleria et al., 2015).

The addition of high-fiber oats that contains β-glucan components in foods enhances their fiber content. This addition aids in reducing bowel transit time which prevents constipation and colorectal cancer (Davidson et al., 1991). β-glucan has the ability to lower cholesterol. Various methods for cholesterol reduction have been proposed such as the consumption or removal of cholesterol precursor being widely accepted (Davidson et al., 1991). According to Pomeroy et al. (2001), oat β-glucan is very useful in lowering blood triglyceride, total cholesterol, and LDL levels. The consumption of dietary fiber including β-glucan is associated with the prevention of stomach

disorders. This is due to the increasing of fecal bulk and higher intestinal viscosity (Wang et al., 2002).

β-glucan has high viscosity which contributes to the suppression of appetite making it ideal for weight reduction. Mostly, viscosity depends on the extracted β-glucan's molecular weight. Due to more connections in the chain, the greater molecular weights produce increased viscosity and gelling qualities (Volikakis et al., 2004). Additional parameters affecting the viscous nature of β-glucan in solutions include solubility and concentration (Burkus et al., 2005).

β-Glucan shows promise as a prebiotic ingredient in food products. As the demand for probiotic and prebiotic foods rises the supermarkets worldwide offer a growing range of options like fresh milk, yogurts, kefir, and even fruit juices (Tomasik et al., 2003). This synergy between prebiotics and probiotics is key for gut health (Warrand et al., 2006). Specific prebiotics are fermented by good bacteria in the large intestine, creating short-chain fatty acids (SCFAs), vitamins, butyrate, and other vital nutrients for the gut (Tomasik et al., 2003; Warrand et al., 2006).

2.1.5 Emulsions and Mechanisms of Instability

Emulsions represent a distinct category of dispersed systems that are characterized by the presence of two liquids that do not naturally mix (Morlat-Therias et al., 2005). Within this system, one liquid forms droplet (disperse phase) which are then dispersed within another liquid, termed as continuous phase. Emulsions can be classified into several types, including oil-in-water (O/W), water-in-oil (W/O), and oil-in-oil (O/O). To achieve the dispersion of two immiscible liquids a third component known as emulsifier is essential. The selection of the emulsifier plays a pivotal role not only in initiating the emulsion formation but also in ensuring its long-term stability (Alexandre & Dubois, 2000).

Emulsions can break down during storage. The figure 3 Illustrates various breakdown mechanisms in emulsions. Some of which are described in the following section in detail. This breakdown depends on several factors. These factors include droplet size and density compared to the surrounding liquid (creaming/sedimentation). They also include the attraction causing clumps (flocculation). Droplet solubility affects their growth (Ostwald ripening). Additionally, the surrounding film's stability impacts merging (coalescence).

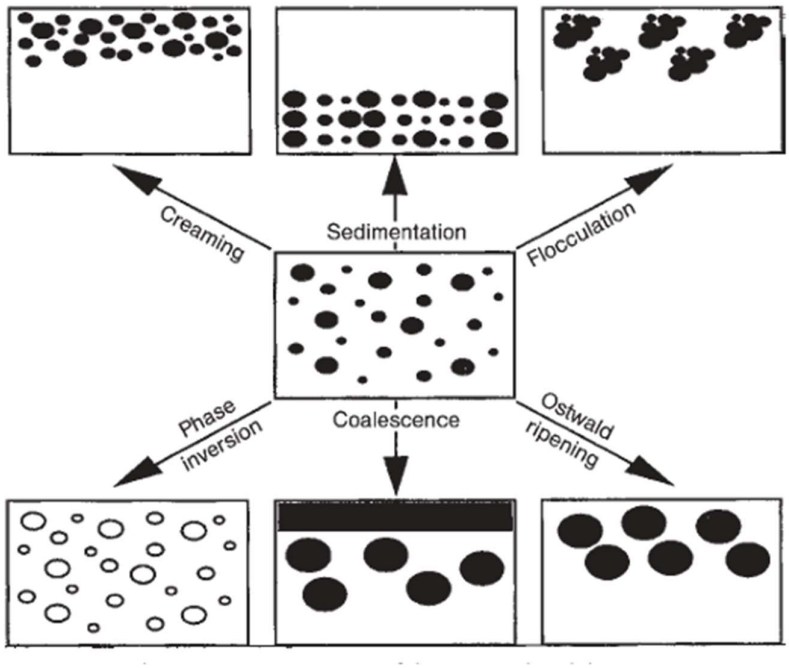

Figure 3; Depiction of Destabilization Mechanisms in Emulsions (Tadros, 2009).

Creaming and Sedimentation

When external forces like gravity exceed the thermal motion of droplets, creaming and sedimentation take place. It causes the system to have a concentration gradient. Then, larger droplets travel faster to the bottom of the container (if their density is greater than that of the medium) or to the top (if their density is lower than that of the surrounding medium). In extreme situations, the continuous liquid phase may occupy the remaining volume while droplets form a densely packed array at the top or bottom (McClements, 2007).

In emulsions resistant to creaming the droplets remain stationary despite gravitational forces. This indicates the elastic behavior rather than viscous fluidity. Rheology is employed to quantify this elasticity. Many industrial emulsions exhibit macroscopic elastic behavior over time and often due to thickeners like xanthan gum.

Flocculation

Flocculation describes the association of two or more droplets while preserving their individual integrities. Usually, it occurs when long-range repulsive interactions between droplets are outweighed by attractive interactions, but not short-range repulsive ones. This balance keeps the droplets closer without merging (coalescing). The degree and kind of flocculation greatly affects the macroscopic characteristics of emulsions, such as appearance, rheology, and sensory perception (McClements, 2005).

While the flocculation of droplets generally detracts from emulsion quality there are some exceptions. In dilute emulsions like soft drinks, flocculation increases particle size. This hastens gravitational separation and thereby diminishes shelf-life (Chanamai et al., 2000). Additionally, flocculation heightens emulsion viscosity which sometimes leads to gel formation in concentrated emulsions (Demetriades & McClements, 1998). However, certain food products require low viscosity which makes flocculation undesirable. In contrast, controlled flocculation can yield desirable textural characteristics in other products (Parker et al., 1994).

Ostwald Ripening (Disproportionation)

Large droplets develop at the expense of smaller ones as a result of mass transfer of dispersed phase material from one droplet to another through the continuous phase, a phenomenon known as Ostwald ripening (OR) (Kabalnov, 2001). Large droplets are shown growing at the expense of smaller ones in Figure 4, which depicts the Ostwald ripening. The disperse phase's molecular diffusion between the droplets is the cause of Ostwald ripening. The solubility of a material increases inside a spherical droplet as its size decreases, which is what propels this process (Kabalnov, 2001).

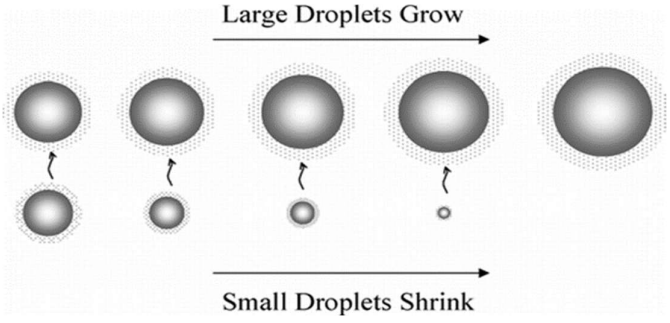

Figure 4; The Ostwald ripening process is illustrated in a schematic diagram.

Coalescence

The process by which two or more liquid droplets combine to form a single, bigger droplet is known as coalescence. It accelerates creaming or sedimentation due to increased size. Furthermore, because larger droplets scatter light less effectively, it can change the appearance of the emulsion. This alteration results in reduced turbidity and more intense coloration (McClements, 2005). In O/W emulsions, coalescence causes oiling off which forms an oil layer atop the material. In W/O emulsions, coalescence causes water accumulation at the bottom.

In actuality, collisions between freely moving droplets rarely result in coalescence in food emulsions. Instead, it typically arises after prolonged contact periods (for instance, in cream layers, or flocs) especially under high stress (van Aken & van Vliet, 2002). The presence of large droplets from coalescence can significantly affect coalescence characterization. If extensive coalescence forms very large droplets or causes oiling off, a selected sample for analysis may not represent the entire emulsion. For instance, sampling from the bottom may yield relatively small measured droplet sizes as larger droplets have migrated to the top and are excluded from collection.

Phase Inversion

The term "phase inversion" describes the change in a system's composition from an oil-in-water to a water-in-oil emulsion, or the opposite. This transformation plays a crucial role in producing various food items like butter and margarine. An O/W emulsion is changed into a W/O emulsion in these goods (McClements, 2005; Walstra, 2003). Nevertheless, phase inversion is unfavorable in other food products. Changes in emulsion composition or environment trigger phase inversion. Variables include temperature, mechanical agitation, emulsifier type and concentration, solvent type, additives, and disperse phase volume fraction (Kabalnov, 1998; McClements, 2005). Phase inversion is not possible for every emulsion. A kinetically stable state can only be maintained by specific types both before and after the process. Agitation is often necessary throughout phase inversion to prevent the emulsion from separating into its constituent phases post-transition.

Chemical and Biochemical stability

Chemical and biochemical reactions present additional challenges to the stability of food emulsions beyond physical instability. Lipid oxidation results in the production of undesirable compounds and off-flavors. Some of

these compounds possess surface-active properties that directly impacts emulsion stability at the interfacial level. Enzyme hydrolysis is utilized to enhance stability by cross-linking interfacial layers surrounding lipid droplets. This is another area of interest in biochemical stability. Colors and flavors added to emulsions for sensory enhancement are susceptible to degradation. Prolonged exposure to light or adverse pH conditions causes this deterioration (McClements, 2016).

2.1.6 How to stabilize Emulsions

Despite their kinetic stability, emulsions tend to separate due to their thermodynamic instability. The tension that exists between the continuous phase and the dispersed phase is the cause of this instability. For example, oil droplets in water create a positive interfacial energy term due to high interfacial tension (typically 1-10 mN/m). As a result, the system minimizes the interfacial area, which lowers the region at which oil and water come into contact (Capek, 2004).

To achieve kinetic stability, emulsions employ several strategies. Firstly, manipulating the size of oil droplets helps. Smaller droplets, with a larger surface area to volume ratio, are inherently more stable due to reduced gravitational forces (creaming) (McClements, 2005). Secondly, surface-active emulsifiers play a crucial role. These molecules, being amphiphilic, adsorb at the oil-water interface, forming a protective barrier against droplet coalescence (Tadros, 2009). Finally, the addition of stabilizing or thickening agents, like polymers or starches, further enhances stability (Dickinson, 1992).

It is important to note that the focus of this project is to investigate the use of β-glucans, as a stabilizing agent in oil-in-water emulsions. β-glucans are gaining significant interest due to their potential advantages, such as being natural, biocompatible, and offering additional functional properties.

From Immiscible to Uniform Emulsion: Homogenization

Emulsion formation is a two-step process achieved through primary and secondary homogenization. Primary homogenization involves the mixing of ingredients to form a coarse emulsion with a broad size distribution. This initial step lays the groundwork for the final emulsion properties. Secondary homogenization, often employing high-shear devices to further refine the emulsion by reducing droplet size and achieving a narrower size distribution for improved stability and functionality. This stepwise approach allows for

precise control over the final characteristics of the emulsion (McClements, 2016).

Achieving stable emulsions in food production presents significant hurdles. High-energy input is typically required. Additionally, careful control over the emulsification process is crucial for ensuring the required quality of products. The size distribution of the distributed droplets has a significant impact on sensory elements such as color and mouthfeel (Chung & McClements, 2014; McClements, 2016).

The thermodynamic instability of emulsions poses a key barrier in their creation. Two essential components are needed to combine two immiscible liquids: (a) an emulsifier, which lowers interfacial tension and stops droplet merging, and (b) enough energy to offset the interfacial energy increase that happens during emulsification. Industrial food processing frequently uses high-energy methods, in which extra energy is added to accelerate the process. The food sector is the primary use for these high-energy emulsification machines, also referred to as homogenizers because of their capacity to improve dispersion uniformity (Håkansson, 2019).

The high-energy emulsification techniques are widely used in the food sector because they can enable huge volume outputs surpassing 50,000 liters per hour to be produced continuously. But this advantage comes at the cost of an extremely low thermodynamic efficiency. A 5% oil-in-water emulsion with 1 μm droplets can be produced with theoretically little interfacial energy—about 2 kJ/m3. According to Mohr (1987), the most popular high-energy technologies use more than a thousand times that amount.

A wide range of high-energy emulsification tools are used in food science. In food production, rotor-stator mixers (RSM) and high-pressure homogenizers (HPH) are the most commonly used emulsification techniques. A high-pressure homogenizer's general design includes both the high-pressure dispersion unit and a high-pressure pump, often a one to three-piston plunger pump powered by electricity or pneumatics. Different configurations of high-pressure dispersion units are available, such as axially flown-through nozzle aggregates, counter jet dispergators, and radial diffusers (Schultz et al., 2004).

The Panda plus 2000 HPH represents a pinnacle in the field of high-pressure homogenization, distinguished by its robust Compression Block and Homogenizing Valve mechanisms (Figure 5). The figure 5 shows the picture of the Panda plus 2000 HPH. It is Constructed from high-grade Super Duplex stainless steel, the Compression Block ensures optimal stress resistance,

critical for sustained operation under high pressures. Noteworthy features such as the solid ceramic pumping piston and specialized packing design eliminate the requirement for lubrication water, thereby augmenting operational efficiency. The Homogenizing Valve, incorporating reversible wear parts and manual pressure adjustment capabilities, offers adaptability across a spectrum of applications (PandaPlus manual, 2024).

HPH forces liquids through a tiny nozzle at high pressure (10-500 MPa, up to 350 MPa in some cases). This creates intense shear stress. Inside the HPH, a pre-mix accelerates through a narrow gap, causing turbulence as it exits. This forceful shearing breaks up the dispersed phase into smaller droplets. HPH offers precise control over droplet size by adjusting pressure and energy input. This makes it ideal for stabilizing bio-oil emulsions. Additionally, high pressure can disrupt weak bonds in large molecules, allowing control over physical and chemical changes that enhance emulsion stability.

HPH also utilizes high-pressure gradient turbulence. When highly pressurized suspensions depressurize, strong shearing forces are generated. This can break apart cell walls and sludge flocs (at pressures like 900 bar), releasing internal substances that improve biodegradation. Studies show increasing pressure and homogenization cycles improves solubilization, highlighting HPH's versatility (Yong et al., 2017).

Figure 5; Gea Niro Soavi Panda Plus 2000 HPH

Soy Lecithin; An Emulsifier

In addition to the previously mentioned formation of finer droplets, an emulsifier must be added in order to create a long-lasting emulsion. This component hinders these droplets from recombining after homogenization. The most important stabilizers in any emulsion composition are emulsifiers. The selected emulsifier affects not only stabilizing properties but also the ease of emulsion formation and functional properties of the finished product. Consequently, selecting a suitable emulsifier is crucial for formulating emulsion-based products. Amphiphilic compounds with both hydrophilic and hydrophobic groups within the same structure are commonly known as emulsifiers. Small molecule surfactants, proteins, phospholipids, polysaccharides, and other surface-active polymers are a few examples (McClements & Jafari, 2018).

Once big droplets break up into smaller ones, it's important to keep them from coalescing inside the homogenizer. The freshly produced droplet surfaces are not completely covered with emulsifier right after a big droplet breaks up. The larger oil-water interfacial area is the cause. The degree of surface coverage determines how stable lipid droplets are against coalescence within a homogenization chamber. Stable droplets are the result of strong repulsive forces and enough emulsifier coverage. On the other hand, in the event that the droplets are only partially covered by the available emulsifier, coalescence may occur upon impact. Therefore, it is essential that before the lipid droplets collide with their neighbors, their surfaces be fully covered in emulsifier molecules.

A naturally occurring combination of phospholipids, mostly phosphatidylcholine and phosphatidylethanolamine, is called soy lecithin. These phospholipids are a waste product from the manufacturing of soybean oil. A degumming step separates them from the oil using water precipitation, forming a liquid crystalline phase. Further processing with acetone is required to produce oil-free, granular Soy Lecithin with a purity of around 95-97% (List, 2015). Due to its amphiphilic nature, soy lecithin can concentrate in emulsions at the oil-water interface.

This action reduces interfacial tension, facilitating the breakdown of oil droplets into smaller sizes. In food applications, the typical usage level of soybean Soy Lecithin varies depending on the type of emulsion. For water-in-oil emulsions, the range is 1-5%, while oil-in-water emulsions typically require 5-10% based on the oil content (List, 2015). According to studies, soy lecithin provides oil-in-water emulsions with better stability against creaming at a pH of about 6.2 (Comas et al., 2006).

The expansion of soy farming, particularly in South America, is a major driver of deforestation, reducing biodiversity, disrupting water cycles, and contributing to climate change. Additionally, soybean cultivation relies on fertilizers, pesticides, and herbicides, which can contaminate waterways and harm aquatic ecosystems.

To achieve a truly eco-friendly product, sustainable soy practices are crucial. These include reducing chemical use, improving soil health, and using certified sustainable soy. While soy lecithin itself offers some environmental benefits (biodegradable, lower carbon footprint than some alternatives), addressing these sustainability concerns in soy production is essential.

Gluten Free (GF) PromOat®-Oat β-Glucan

β-glucans are categorized as hydrocolloids due to their affinity for water. When hydrocolloids are dissolved in water, they produce gels and/or viscous dispersions. Hydrocolloids directly affect the viscosity of the product (flow behavior) and texture by altering the rheology of the food system. This is important for the food product's long-term stability as well as its sensory qualities. Due to the physical entanglement of conformational disordered random coils, hydrocolloids alter a solution's viscosity (Saha and Bhattacharya, 2010). This causes an increase in flow resistance when shearing forces are applied. Viscosity is directly influenced by a hydrocolloid's concentration in a solution; at low concentrations (dilute solutions), the viscosity increases linearly and exhibits Newtonian fluid behavior. As the concentration increases and the molecules start to overlap, the viscosity increases significantly due to the enhanced molecular entanglement. This concentration, known as the critical overlap concentration (c*), is determined by the hydrocolloid's intrinsic viscosity or molecular volume occupancy (Goff and Guo, 2019). Shear thinning behavior of the dispersion is observed above c* (Saha and Bhattacharya, 2010).

Finding c* helps us understand β-glucans in solution better, which will help us develop more successful product applications (Burkus, 2003). Molecular weight, extraction source, temperature, and pH are some additional factors that influence the viscosity of β-glucan solutions. Solutions containing β-glucan exhibit stability within the pH range of 2-10. The stability decreases with the increase in temperature, with the exception of temperatures over 75°C, which cause a reduction in viscosity. Increased molecular weight according to Ahmad and Kaleem (2018), β-glucans are believed to produce solutions with a higher viscosity relative to their molecular weight.

Concerns about the function of β-glucans as an ingredient have also been sparked by the gelling abilities of β-glucan solutions. Hydrogen bonds unite water-trapping β (1-3) connected cellotriosyl units to form junction zones. A gel network with cluster-cluster aggregation is formed when the strands aggregate with one another (Brummer et al., 2014). Gel-forming potential of β-glucans is related to the proportion of cellotriosyl to cellotetraosyl subunits in the cereal structure. Compared to barley and oat, wheat β-glucans have a higher likelihood of forming gels due to their relative percentages of cellotriosyl to cellotetraosyl units, which are 4, 3, and 2, as reported by Lazarides et al. (2003). Gels with longer chain segments exhibit superior order and interchain connections when made using β-glucans with higher molecular weight. Low molecular weight β-glucans, on the other hand, show a faster gelation process. Greater molecular weight gels therefore have a greater melting point (Lazaridou et al., 2003).

2.1.7 Packaging Evaluation of Gf PromOat®

This thesis also delves into the packaging and labelling of GF PromOat®, a potential source of health benefits like cholesterol reduction and blood sugar control. However, proper packaging and storage is crucial for its effectiveness. This research assesses GF PromOat®'s packaging in terms of preserving product integrity and facilitating portion control. Additionally, the labelling is evaluated for clear communication of health claims and compliance with relevant regulations. The findings aim to provide GF PromOat® with valuable insights to optimize their packaging and labelling strategies, ultimately enhancing user experience and ensuring consumers receive the intended health benefits.

3 Methodology

This chapter outlines the research methodology, covering the materials used and detailed methods for sample preparation and evaluation. It describes the determination of the critical overlap concentration (C) of Gluten Free PromOat®, including viscosity measurements and intrinsic viscosity evaluation. The viscosity-shear profile of GF PromOat® is also examined. Additionally, the chapter evaluates the role of different ingredients in oil-in-water emulsions and assesses emulsion stability using the Turbiscan LAB Analyzer and viscosity measurements.*

3.1 Materials

Gluten Free PromOat® was provided by Lantmännen as a powder. GF PromOat® powder provided the base. When the β-glucans were taken out, the dry basis content was 30.4% ± 2.0%, with 3.5% of protein, 6.5% of fat, 42.5% of carbohydrates, and 6.0% of fiber. As per Lanthmännen, the molecular weight was within the range of 1,03 MDa. Rapeseed oil (batch REV14092, Zeta Fernando Di Luca) was chosen as the oil phase. To achieve a stable emulsion, Bungemax Soy Lecithin (lot number CSL 02/22 DOL 01)

was incorporated as the emulsifier. A premade 0.05 M buffer solution comprising sodium hydroxide, lactic acid, and acetic acid with a regulated pH of 6.2 was also utilized. The buffer solution was supplied by EMD Millipore Corporation (Darmstadt, Germany).

3.2 Methods

The evaluation methods described in section 3.2 are flexible, allowing for adjustments based on the project's specific requirements. Below is a general explanation to provide a clear understanding of these methods.

3.2.1 Sample Preparation

The GF PromOat® powder was gradually incorporated into a well-stirred buffer solution (pH 6.2) using a high-shear mixer (T 25 Ultra-Turrax, IKA-Werke GmbH & Co. KG, Germany) for five minutes at 24,000 rpm using the S 25 N-18 G dispersing tool. This step ensures proper hydration and dispersion of the beta-glucan particles. Next, the rapeseed oil and Soy Lecithin were added to the previously prepared beta-glucan dispersion. After that, the mixture was homogenized once more for five minutes at 24,000 rpm using the Ultra-Turrax. This step facilitates the initial emulsification of the oil phase within the beta-glucan dispersion. Using a PandaPLUS 2000 (GEA Niro Soavi S.p.A., Italy), the produced emulsions were then treated with High Pressure Homogenization (HPH).

3.2.2 Sample evaluation

To explore how different ingredients affect emulsions, various formulations were created as specified in the Appendix A. The apparent viscosity, oil droplet size, and visual inspection of the samples were then used to assess them.

Viscosity Measurement

Every sample's apparent viscosity was assessed. The viscosity of the emulsions was determined using a rheometer (Kinexus Pro+) featuring a concentric cylinder shape that is smooth. This geometry consisted of a PC25 C0003SS cup and a C25 SC0053SS bob. The rheometer was initially calibrated using a ten percent (mass/mass) sucrose solution at 25°C and a

known viscosity of 1.25 mPa.s to guarantee measurement accuracy. For each emulsion sample, 16 ml was loaded into the rheometer's measurement chamber. Next, ten measurements of each sample's apparent viscosity were made at a regulated temperature of 25°C and a constant shear rate. This approach helps account for any minor variations and provides a reliable average viscosity value for each emulsion. Finally, the average viscosity for each sample was calculated.

Measurement of Droplet Size

Using a Mastersizer 2000 laser diffraction particle size analyzer, the size of the oil droplets was determined both before and after homogenization. Refractive indices of 1.47 for rapeseed oil and 1.33 for water were employed. As directed by the instruction's manual, 3-5 drops of undiluted emulsion were added to the dispersant (deionized water) until the laser obturation reached 2.0% ± 0.2%.

Visual Inspection

The samples were visually inspected for any signs of separation or instability, such as creaming, flocculation, or sedimentation. Photographs were taken of each sample at the start of the inspection process and at regular intervals throughout the storage period to document any visual changes over time. The samples were examined under consistent lighting conditions to ensure uniformity in the assessment. Additional protocols for visual inspection included storing samples at room temperature (25°C).

3.3 Calculating the critical overlap concentration (C*) of GF PromOat®

3.3.1 Viscosity Measurements

Five different solutions of GF PromOat® with increasing concentrations (0.02%, 0.04%, 0.06%, 0.08%, and 0.10%) were prepared in ten percent (mass/mass) sucrose solution. The apparent viscosity of each solution was measured following the procedure described in section 3.2.1. The only variation in the measurement was the change in shear rate, as the test was performed at a constant shear rate of 50 s^{-1}.

3.3.2 Intrinsic viscosity and the C* evaluation

To determine the intrinsic viscosity the reduced viscosity for each sample was calculated using Equation 1.

$$\eta red = (\eta - \eta 0) / (\eta 0\ c) \qquad (1)$$

This equation relates the reduced viscosity (ηred) to the sample viscosity (η), solvent viscosity (η0, measured with the 10% sucrose solution), and sample concentration (c in g/mL).

By applying linear regression to extrapolate the reduced viscosity values to zero concentration, intrinsic viscosity (η) was found.

Finally, the critical overlap concentration (c*), which signifies the point where entangled polymer chains begin to interact, was determined using Equation 2 (Burkus, 2003).

$$c* \approx 2.5/[\eta] \qquad (2)$$

η= Intrinsic viscosity

This equation relates intrinsic viscosity (η) with c*.

3.3.3 Verifying C* calculation

The rheological behavior of various GF PromOat® solutions was examined in order to confirm the accuracy of results obtained in the c* calculation section. The different GF PromOat® concentrations as shown in the table 1 were dissolved in water and apparent viscosities were measured as described in section 3.2.1.

GF PromOat® (%)	GF PromOat® (g/ml)
1.00	0.01
2.00	0.02
3.49 (c*)	0.0349
4.00	0.04
5.00	0.05

Table 1; GF PromOat® concentrations used for c* verification

3.4 GF PromOat® Viscosity-Shear Profile Measurement

This experiment investigated the viscosity of GF PromOat® solution containing 3.49% of Gf PromOat® at shear rate of 0.1 s-1 to 5000 s-1. A rheometer described in the section 3.2.2 was used. The graph containing apparent viscosity of solution at different shear rates was obtained at controlled temperature of 25°C.

3.5 Evaluating Role of Different Ingredients in Oil-in-Water Emulsion

The primary aim of this experiment was to identify the optimal concentrations and combinations of Soy Lecithin and GF PromOat® that enhance the stability and viscosity of emulsions.

3.5.1 Sample Preparation

The samples were prepared with different composition of ingredients. In Trial 1, emulsions only with varying concentrations of Soy Lecithin (4 g, 4.5 g, 5 g, 5.5 g, and 6 g) were prepared. The Gf PromOat® was not used in Trial 1. Trial 2 focused on GF PromOat® and involved incorporating different concentrations based on a critical overlap concentration (c*) calculated in section 3.3.2. Trial 3 combined Soy Lecithin and GF PromOat® to assess any synergistic effects. New formulations combined constant amount of Soy Lecithin (5 g) with varying concentrations of GF PromOat®. Trial 4 investigated the optimal balance between Soy Lecithin and GF PromOat® concentration by subjecting them to HPH at 300 bar. New formulations for various GF PromOat® concentrations were prepared, with different Soy Lecithin concentrations corresponding to 20%, 40%, 60%, 80%, and 100% of the GF PromOat® amount. To understand the impact of number of circulations during HPH, Emulsion A was prepared (Table 1). It underwent three homogenization treatments at 300 bar: single (1x), double (2x), and triple (3x) recirculation.

3.5.2 Sample evaluation

To explore the impact of different ingredients on emulsions, various evaluations were made. First, the samples underwent visual inspection for signs of instability, such as creaming or separation, as detailed in section 3.2.2. Additionally, the apparent viscosity of each sample was measured according to the procedures in section 3.2.2. The oil droplet size was also measured immediately after homogenization.

3.6 Assessing Emulsion Stability

3.6.1 Sample Preparation

Emulsion A was made in order to assess GF PromOat®'s performance as an agent of stabilization in oil-in-water emulsions. The composition of Emulsion A is given in table 2. The emulsion preparation followed the specified method as described in section 3.2.1. However, the pressure during HPH differed, set at 100 bar, 300 bar, and 700 bar. Additionally, the emulsion underwent three recirculations through the equipment.

Emulsion A			
Soy Lecithin	GF PromOat® (g)	Rapeseed oil (g)	Buffer solution (g)
0.523(15%)	3.49	25	71

Table 2; Emulsion selected for stability testing

3.6.2 Stability Testing with Turbiscan LAB Analyzer

To assess emulsion stability, samples were prepared for Turbiscan LAB analysis. Glass cells were meticulously cleaned to avoid result interference. Each emulsion (20 mL) was carefully pipetted into a cell, ensuring the tip reached the bottom to prevent air bubbles. Any meniscus at the top was gently tapped against the holder for a uniform surface. The cells were then sealed with specialized black caps and rubber seals to prevent leakage. The

samples were labeled for identification and kept for 30 days at room temperature. Importantly, the samples remained undisturbed after the initial measurement to preserve the droplet distribution for accurate stability evaluation.

3.6.3 Emulsion Viscosity Measurement

The method outlined in section 3.2.2 was used to measure the emulsions' viscosity.

3.6.4 Determination of the Stability

To determine the stability of the emulsions over time, the Turbiscan LAB Analyzer was used to create droplet profiles. Immediately after sample preparation, the glass cells were scanned using the instrument's single scan setting at a controlled temperature of 25°C. The scanning schedule employed a high initial frequency: measurements were taken every 2 hours for the first 6 hours and once after every 24 hours for the first week. This allows for capturing any rapid changes that might occur during the early stages of emulsion formation. Following this initial period, measurements were taken once every week for next 4 weeks. This comprehensive scanning approach aims to detect potential instability phenomena at all stages of the experiment, from the initial formation to the long-term storage.

The Turbiscan LAB Analyzer serves a dual purpose beyond assessing overall stability—it facilitates the identification of creaming rate. By conducting backscattering measurements, expressed in percentage, it monitors changes in backscattering across sample sections, indicative of specific instabilities, as outlined in the accompanying table 3. Essentially, this feature tracks variations in light scattering (turbidity) throughout the sample's height. This data enables the determination of creaming rates for individual emulsions.

ΔBS	Bottom	Middle	Top	Instability
Case 1	⇑	-	⇓	Sedimentation
Case 2	⇓	-	⇑	Creaming
Case 3	⇓	⇓	⇓	Flocculation or coalescence

Table 3; Instability phenomena correlated to the backscattering signal (Turbiscan, 2024).

4 Results and Discussion

This chapter presents and analyzes the study's findings, including the critical overlap concentration (C) determination for Gluten Free PromOat®, viscosity-shear profile, and emulsion stability. It discusses the effectiveness of ingredient combinations in stabilizing emulsions and interprets results from stability testing using the Turbiscan LAB Analyzer, providing insights for practical applications.*

4.1 Calculating the critical overlap concentration (C*) of GF PromOat®

4.1.1 Measurements of GF PromOat® Solutions' Apparent Viscosity

Table 4 shows the results of apparent viscosity. It can be seen that the values increased with increasing GF PromOat®. A 10% m/m sucrose solution had an average apparent viscosity of 1.25 ± 0.04 mPa.s. The apparent viscosity increased linearly and the addition of 0.10 g/mL GF PromOat® (1.0%) resulted in an apparent viscosity of 1.80 ± 0.03 mPa.s, representing a 44% increase compared to the sucrose solution alone.

Concentration (g/ mL)	Apparent viscosity (mPa.s)
0.0000	1.25 ± 0.04
0.0002	1.27± 0.03
0.0004	1.39± 0.04
0.0006	1.45± 0.04
0.0008	1.76± 0.04
0.0010	1.80± 0.03

Table 4: Apparent Viscosity of GF PromOat® Solutions

4.1.2 finding out the critical overlap concentration and intrinsic viscosity.

To determine the reduced viscosity, Equation 1 was applied to each value listed in Table 4. The resulting data is presented in Table 5. Furthermore, the obtained reduced viscosity values and the varying concentrations of GF PromOat® were utilized to establish a correlation, as depicted in Figure 6.

Apparent Viscosity (**η**) of sample	concentration of the sample in g/mL (c)	viscosity of the 10% m/m sucrose solution ($\eta 0$)	Reduced viscosity (ηred)
1.25 ± 0.04	0.0000	1.25	0
1.27± 0.03	0.0002	1.25	80
1.39± 0.04	0.0004	1.25	140
1.45± 0.04	0.0006	1.25	267
1.76± 0.04	0.0008	1.25	510
1.80± 0.03	0.0010	1.25	520

Table 5; Calculated Reduced viscosities.

To determine the intrinsic viscosity, the reduced viscosity was linearly extrapolated to zero concentration. The value, which represents the molecular occupancy of each molecule in the solvent, is **71.6 ml/g** as shown in the figure 6.

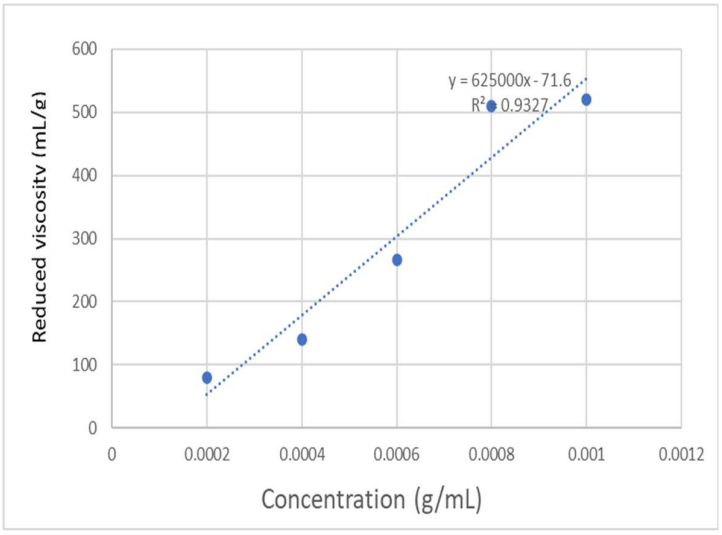

Figure 6; Reduced Viscosity plotted against the concentration of GF PromOat® in the sample.

The equation 2 estimates the c*, which is the level of concentration at which colloids or dissolved polymers start to entangle and overlap. According to the calculation, GF PromOat® should have a c* of **0.0349 g/mL**, or 3.49% (m/m) of the solution and 0.01047 g/mL in relation to the 30% β-glucan percentage in the powder.

4.1.3 Verifying C* calculation

In the figure 7, the provided graph depicts the apparent viscosity of GF PromOat® as a function of concentration, exhibiting two distinct linear regimes characteristic of disordered polysaccharide solutions.

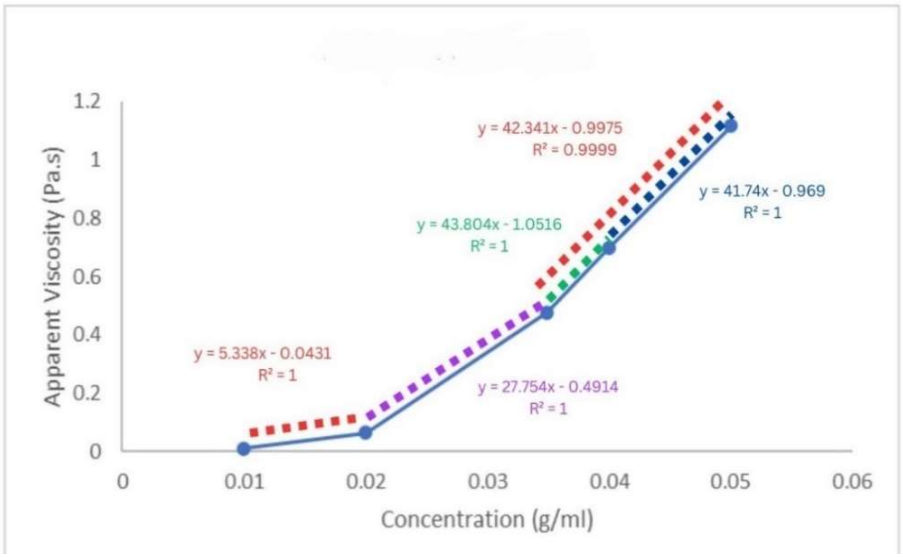

Figure 7; Relation between Apparent viscosity and GF PromOat® Concentration

In order to understand the role of GF PromOat® on the apparent viscosity, consider two regions plotted in red color. At lower concentrations, the first region demonstrates a lesser increase in apparent viscosity with increasing concentration. This can be attributed to the well-separated nature of individual polymer molecules in dilute solution. As concentration increases, the viscosity also increases, but in a gradual manner.

The second region, observed at higher concentrations, showcases a significantly steeper increase in apparent viscosity. This phenomenon coincides with reaching the critical overlap concentration (c*) of 3.49%. At this crucial point, the polymer chains become entangled, resulting in enhanced chain-to-chain interactions. These interactions are responsible for the substantial upsurge in viscosity. The slopes of the two linear regions quantify the rate of viscosity change with concentration. The slope in the first region is 5.33, while the slope in the second region exhibits an almost eight-fold increase, reaching 42.3.

The presence of equations for both linear regions on the graph, accompanied by their corresponding R-squared values, allows for a quantitative analysis of the relationship between concentration and apparent viscosity. R-squared, a statistical measure that reflects how well the data aligns with the fitted model, with a value of 1 indicating a perfect fit.

4.2 GF PromOat® Viscosity-Shear Profile Measurement

While random coil polysaccharides typically show a decrease in viscosity (shear thinning) under flow due to molecule alignment, GF PromOat® solutions can behave differently. Their flow properties may exhibit an upward trend. This is because, as mentioned earlier, GF PromOat® can form gels. Through internal interactions or aggregations, these gels form a continuous network of molecules. The observed deviation from the expected shear thinning behavior is caused by this network topology (Salovaara, 2007).

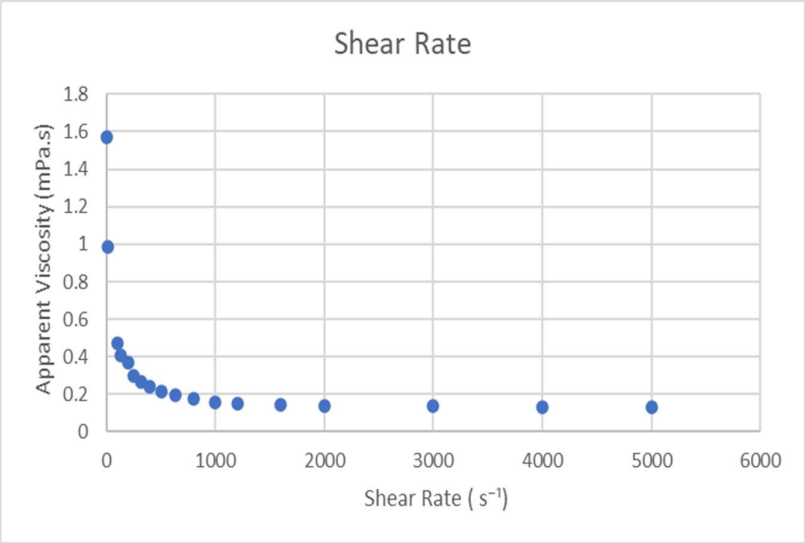

Figure 8; Apparent viscosity as a function of shear rate of emulsion having 3.49% GF PromOat®.

Figure 8 illustrates how the thickness (viscosity) of GF PromOat® changes with increasing force applied (shear rate). At low force, the tangled polymer chains break apart (disentangle) and reconnect (entangle) at similar rates, keeping the solution thick. However, as the force gets stronger, disentanglement outpaces entanglement due to random molecular movement. This rapid disentanglement significantly reduces the thickness, a phenomenon called shear thinning. With even stronger force, the molecules become almost disentangled, resulting in a constant and low viscosity.

The Figure 8 also depicts the possibility of gel formation in GF PromOat®, which could influence its response to force. Gel formation depends on the molecular weight of the β-glucan (higher weight promotes gels), the ratio of

specific sugar structures within the molecule, and storage time (gels can develop over time). However, several factors suggest minimal gel formation in this case. Oat β-glucan, compared to other cereals, is less likely to form gels due to its specific sugar unit ratio. Additionally, the experiment used fresh samples, and gel formation for GF PromOat®'s high molecular weight β-glucan likely would not occur within the short timeframe of the experiment. Therefore, the observed flow behavior in Figure 3 is primarily due to disentanglement and shear thinning of the GF PromOat® solution, with minimal influence from gel formation.

4.3 Evaluating Role of Different Ingredients in Oil-in-Water Emulsion

Trial 1. Role of Soy Lecithin

To explore how different ingredients affect emulsion viscosity, four trial formulations were created in triplicate. Started by investigating the role of Soy Lecithin, a crucial emulsifier. Emulsions were prepared with varying Soy Lecithin concentration as shown in the table 6.

Soy Lecithin (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
4.0	25	71.0
4.5	25	70.5
5.0	25	70.0
5.5	25	69.5
6.0	25	69.0

Table 6; Emulsions with varying amounts of Soy Lecithin

The trial one showed following apparent viscosity trend (figure 9);

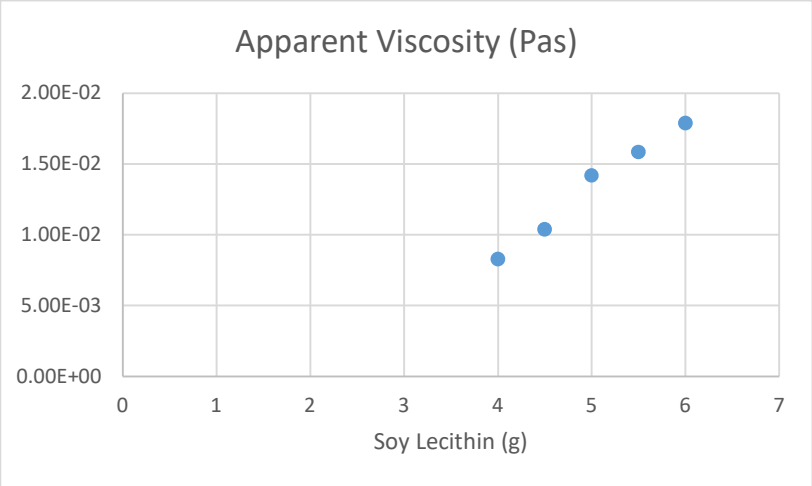

Figure 9; Apparent Viscosity (Pas) of emulsions containing only Soy Lecithin

The current results align with established scientific principles regarding Soy Lecithin's role in emulsions. As expected, the apparent viscosity increased with increasing Soy Lecithin concentration. This can be attributed to the well-documented function of Soy Lecithin molecules. They act as emulsifiers, possessing both hydrophilic (water-loving) and lipophilic (oil-loving) properties. This allows them to position themselves at the oil-water interface, reducing interfacial tension and preventing droplet aggregation. As Soy Lecithin concentration increases, more molecules pack at the interface, creating a thicker layer that hinders droplet movement, resulting in higher viscosity.

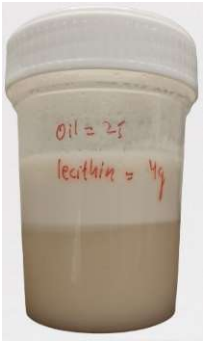

Figure 10; Emulsion with only Soy Lecithin(4g)

In contrast to the expected behavior, the emulsions separated immediately after creation. This could be seen in Figure 10. This suggests that the chosen Soy Lecithin concentration might be outside the optimal range for this particular system. Soy Lecithin is known to improve emulsion stability by reducing interfacial tension. However, scientific research indicates that excessive amounts can have a destabilizing effect (McClements, 2005; Ono et al., 1994).

Two primary mechanisms could explain this observed instability. First, steric repulsion occurs when an excessive amount of Soy Lecithin is incorporated. It causes the molecules at the interface to repel each other due to steric hindrance (McClements, 2005). This repulsive force can disrupt the emulsion's stability and lead to phase separation. Second, phase inversion is a potential issue. Soy Lecithin's function as an emulsifier can shift from oil-in-water (o/w) to water-in-oil (w/o) depending on its concentration relative to other ingredients (Ono et al., 1994). If the Soy Lecithin concentration surpasses the optimal level for a desired o/w emulsion, it can cause the emulsion to invert into a w/o type. These findings highlight the importance of optimizing Soy Lecithin concentration for achieving a stable emulsion and the need for a stabilizing agent such as GF PromOat®.

Trial 2; Role of Gf PromOat®

Next, the focus shifted to analyzing the importance of GF PromOat® in stabilizing the emulsions. Different concentrations of GF PromOat® were incorporated, each calculated as a fraction of a previously determined critical overlap concentration (c*) (Table 7).

Fraction of C*	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.25	0.87	25	74.13
0.50	1.75	25	73.25
0.75	2.62	25	72.38
1	3.49	25	71.50
1.5	5.24	25	69.76

Table 7; Emulsions with varying amount of GF PromOat®

The analysis of GF PromOat®'s influence on emulsion viscosity and stability yielded promising results. It aligned with the research on beta-glucan's network-forming properties (Burkus, 2005). As observed, the emulsions displayed enhanced stability with increasing concentrations of GF PromOat® (up to 3.49 g). This increase is indicated by a delay in separation compared to the sample containing least amount of GF PromOat® (0.87 g) that separated immediately. This aligns with the concept of a critical overlap concentration (C*) for beta-glucans. At concentrations above C*, beta-glucan molecules begin to interact and form a network-like structure. This network can potentially entrap oil droplets within the emulsion, hindering creaming and separation (Tadros, 2009).

The emulsion containing the highest GF PromOat® concentration (5.24 g) exhibited excessive viscosity (figure 11), suggesting potential gelation. This observation highlights the importance of optimizing the concentration for achieving a balance between stability and desired consistency.

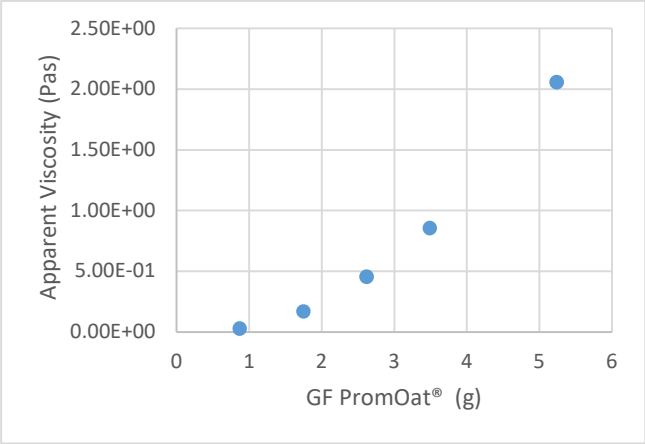

Figure 11; Apparent viscosity of different GF PromOat® emulsions.

The accompanying figure 12 visually confirms the trend. The emulsions with higher GF PromOat® concentrations (from left to right) appear more stable with less separation evident.

Figure 12: Emulsions containing GF PromOat® at concentrations of 0.87 g, 1.75 g, 2.62 g, 3.49 g, and 5.24 g (from left to right).

Overall, these results suggest that GF PromOat®, when used at concentrations near C*, can effectively improve the stability of emulsions. However, a delicate balance exists between achieving stability and encountering excessive viscosity.

Trail 3. Combination of Ingredients

After investigated the individual effects of Soy Lecithin and GF PromOat® on emulsion stability, the next step involved combining them. This aimed to understand any synergistic effects these ingredients might have. New formulations were created that combined a constant amount of Soy Lecithin (5 g) with varying concentrations of GF PromOat®(Table 8).

Soy Lecithin (g)	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
5	0.87	25	69.13
5	1.75	25	68.25
5	2.62	25	67.38
5	3.49	25	66.50
5	5.24	25	64.76

Table 8; Emulsions with varying amount of GF PromOat® but same amount of Soy lecithin.

The third trial was not completely successful. The initial formulations resulted in emulsions with excessively high viscosity. It exceeded the processing capabilities of the high-pressure homogenizer (HPH) which is 0.5 Pas as shown in the figure 13.

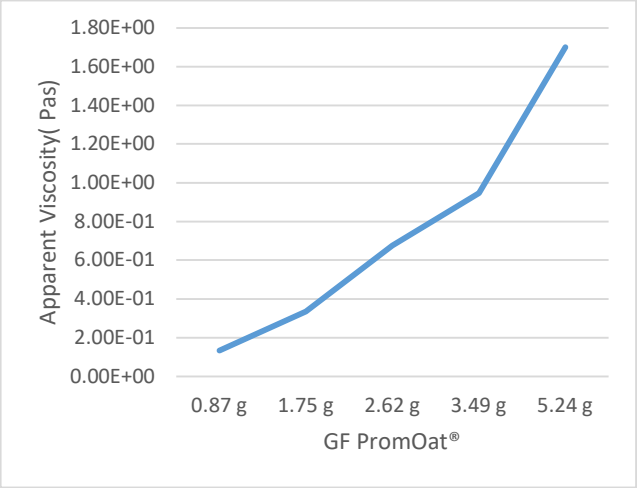

Figure 13; Trend line of apparent viscosity of emulsions containing increasing amount of GF PromOat®.

This high viscosity likely arose from using concentrations of either Soy Lecithin or GF PromOat® that were too high. Soy Lecithin molecules at high concentrations can become densely packed at the oil-water interface, increasing resistance to flow and leading to high viscosity (McClements, 2005). Likewise, an excess of GF PromOat®, has the potential to create a dense network structure at elevated concentrations. This can impede flow and increase viscosity. This underscores the necessity of fine-tuning the concentrations of individual components. It is necessary to strike a harmonious balance between stability and processability. HPH is typically suitable for processing samples with viscosities below 0.5 Pas. The first two samples (with GF PromOat® concentrations of 0.87g and 1.75g) exhibited viscosities within this range. However, these emulsions still separated within an hour, indicating insufficient stability as shown in the figure 14.

Figure 14; Sample containing 0.87g of GF PromOat® showing separation of layers

Trial 4. Optimization of Ingredients

New formulations for various GF PromOat® concentrations (0.87 g, 1.75 g, 2.62 g, 3.49 g, and 5.24 g) were made. For each GF PromOat® level, different Soy Lecithin concentrations i.e. 20%, 40%, 60%, 80%, and 100% of the corresponding GF PromOat® amount were used (table 9). Similar emulsions were prepared for GF PromOat® concentration of 1.75 g, 2.62 g, 3.49 g, and 5.24 g given in the Appendixes A.

Soy Lecithin	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.174g(20% of 0.87)	0.87	25	74
0.348g(40% of 0.87)	0.87	25	73.8
0.622g(60% of 0.87)	0.87	25	73.5
0.7g(80% of 0.87)	0.87	25	73.4
0.87g(100% of 0.87)	0.87	25	73.2

Table 9; Emulsions containing amount of Soy lecithin relative to the GF PromOat® amount

Emulsion with 0.87g GF PromOat®

The emulsions containing 0.87g of GF PromOat® showed a decrease in apparent viscosity after high pressure homogenization as shown in the figure 15. The emulsions showed rapid separation of layers in all samples. It mainly depicts that the lower concentration of GF PromOat® and Soy Lecithin is not suitable for stabilizing the emulsion.

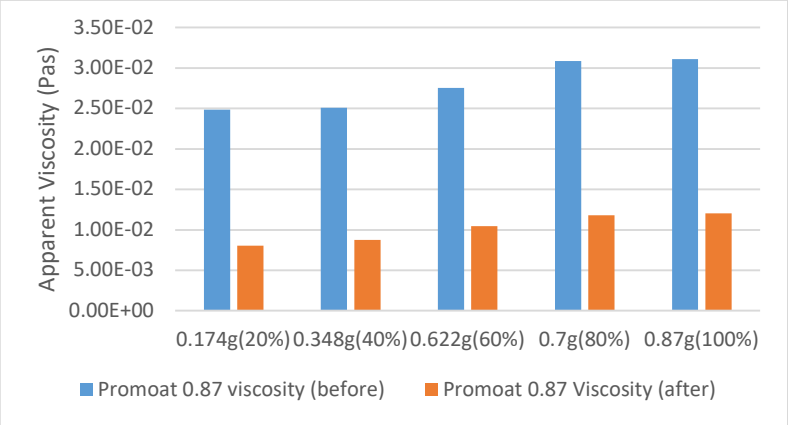

Figure 15; Apparent viscosity of emulsions before and after HPH

1.75g GF PromOat®

In line with prior observations, HPH at 300 bar yielded a decrease in sample viscosity and maintained emulsion stability for one hour. However, droplet size analysis revealed a counterintuitive increase following HPH treatment. This contradicts the established role of HPH in reducing droplet size within emulsions (Figure 16).

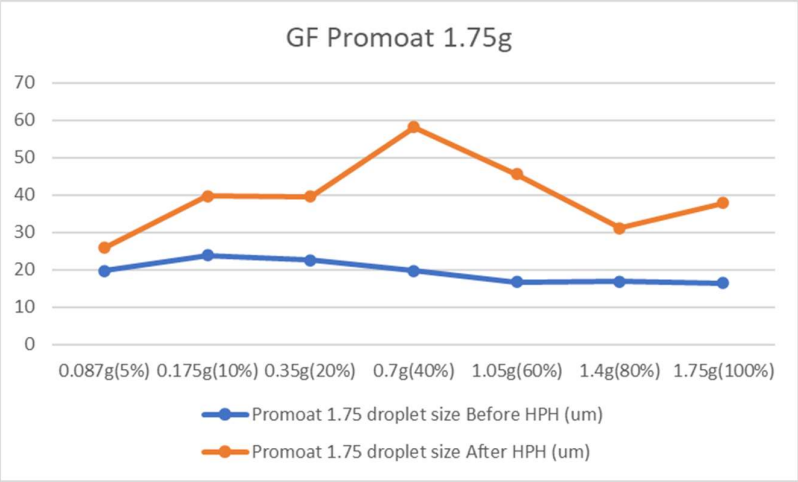

Figure 16; Droplet size before and after HPH (Droplet size on y-axis and Soy lecithin concentration on x-axis).

Two potential explanations are explored. Firstly, the homogenization time may have been excessive despite maintaining a constant pressure as the emulsion were circulated three times through HPH. Research by Floury et al. (2000) suggests that exceeding a certain pressure threshold can lead to destabilization and coalescence even at lower pressures. Secondly, the Soy Lecithin concentration might have been insufficient. McClements et al. (2000) highlight the importance of adequate Soy Lecithin content for effective droplet coating. The employed Soy Lecithin concentrations relative to GF PromOat® (1.75 g) might not have been sufficient to prevent coalescence after HPH.

2.62g GF PromOat®

As anticipated, increasing the GF PromOat® concentration resulted in a corresponding rise in viscosity. Fortunately, the initial emulsions formulated with 5% and 10% Soy Lecithin achieved viscosities that fell within the acceptable range for successful HPH treatment (Table 10).

Soy Lecithin	Viscosity (Pas) (before HPH)	Viscosity (Pas) (after HPH)
0.131g(5%)	0.5201	0.1442
0.262g(10%)	0.5825	0.1861
0.524g(20%)	0.6212	nd
1.048g(40%	0.6431	nd

Table 10; Apparent Viscosity before and after HPH

nd: not determined

However, the effect on droplet size presented a more nuanced scenario. The emulsion containing 5% Soy Lecithin demonstrated an increase in droplet size after HPH treatment (Table 11). This phenomenon suggests the potential occurrence of flocculation. While droplet size is a significant factor in emulsion stability, flocculated droplets can still lead to creaming or sedimentation over time, ultimately causing separation.

Soy Lecithin	Droplet size (µm) (before HPH)	Droplet size (µm) (after HPH)
0.131g(5%)	15.655	45.024
0.262g(10%)	14.596	10.781
0.524g(20%)	18.697	nd
1.048g(40%	17.715	nd

Table 11; Droplet size of emulsion before and after HPH

The emulsion formulated with 10% Soy Lecithin yielded a more promising result. HPH treatment effectively decreased the droplet size (Table 10). Furthermore, this emulsion exhibited better initial stability compared to the one containing 5% Soy Lecithin. This enhanced stability could be attributed to a more thorough coating of the oil droplets by the Soy Lecithin, as highlighted in prior studies. (McClements et al., 2000). However, despite the initial improvement in stability the 10% Soy Lecithin emulsion still exhibited separation after one day. This observation indicates that even with a smaller droplet size and potentially better initial coverage, the Soy Lecithin film may not be robust enough to ensure long-term stability. Research provides a broader perspective on the factors influencing emulsion stability,

highlighting that droplet size is just one piece of the puzzle (Dickinson, 1992).

3.49g (c*) GF PromOat®

Exploring the range of Soy Lecithin concentrations (up to 100%) in the 3.49 g GF PromOat® emulsions yielded interesting results. However, it also presented a challenge. Soy Lecithin concentrations above 15% caused the emulsions to completely gel. Focusing on the more promising range below 15% Soy Lecithin, I observed some intriguing findings. Similar to previous tests, all three emulsions (5%, 10%, and 15% Soy Lecithin) achieved a good consistency but their viscosity exceeded the HPH equipment's capacity (Table 12). Here, the GF PromOat® concentration interacted with the other phases, leading to a thicker mixture.

Soy Lecithin	Viscosity (Pas) (before HPH)	Viscosity (Pas) (after HPH)
0.1745g(5%)	1.177	0.2927
0.349g(10%)	1.137	0.3217
0.523g (15%)	1.259	0.6602

Table 12; Apparent Viscosity before and after HPH

Despite exceeding the HPH limitations, homogenization occurred due to shear thinning behavior. This phenomenon, means certain materials get less viscous under high shear forces. The intense shear during HPH treatment could have temporarily lowered the viscosity, allowing for homogenization. This explains why all three emulsions achieved a reduction in droplet size (Table 13).

Soy Lecithin	Droplet size (µm) (before HPH)	Droplet size (µm) (after HPH)
0.1745g(5%)	25.224	9.877
0.349g(10%)	18.955	6.596
0.523g (15%)	16.106	8.656

Table 13; Droplet size before and after HPH

The stability of the emulsions also varied significantly. The emulsion with the lowest Soy Lecithin content (5%) separated the fastest, after just a few hours. The 10% Soy Lecithin emulsion fared better initially, showing stability for a week before separating. However, the emulsion containing the

highest concentration (15% Soy Lecithin) demonstrated the most promising results, exhibiting the best long-term stability. This difference in stability likely relates to the amount of Soy Lecithin and its ability to coat the oil droplets. Research discusses this concept and suggested that a higher Soy Lecithin concentration can form a more robust coating around the droplets, preventing interactions and ultimately leading to better long-term stability (McClements et al., 2000), as observed in the 15% Soy Lecithin emulsion.

5.24g GF PromOat®

All the samples formulated with 5.24 g of GF PromOat® exhibited complete gelation as shown in the figure 17. This indicates that the chosen concentration exceeded a critical limit (c*). In the context of gelling agents like GF PromOat®, c* represents the minimum concentration required for the formation of a three-dimensional network throughout the mixture. Essentially, below c*, the GF PromOat® molecules are dispersed but don't form a gel. However, at concentrations exceeding c*, these molecules interact and entangle, creating a continuous network that traps water and other ingredients, resulting in the observed gel-like consistency.

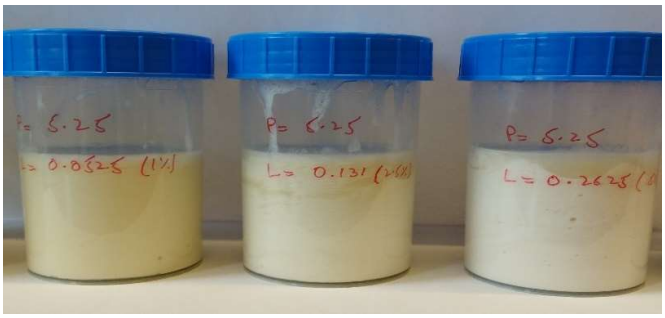

Figure 17; Emulsions showing gelation after addition of 5.24g of GF PromOat®

We know the c* for this system is around 3.49 g. Previous experiments have demonstrated successful emulsions at this concentration. However, the higher concentration of 5.24 g surpassed this threshold. This led to gelation in all samples.

Effect of Number of Circulations

The emulsion A (Table 14) underwent three distinct homogenization treatments during HPH. Firstly, a single circulation (1x) then two (2x) and three (3x) circulations keeping the pressure at 300 bar. The experiment aimed to assess the influence of circulation number during high-pressure

homogenization (HPH) on the particle size, apparent viscosity, and stability of emulsions.

	Emulsion A		
Soy Lecithin	GF PromOat® (g)	Rapeseed oil (g)	Buffer pH 6.2 (g)
0.523(15%)	3.49	25	71

Table 14; Emulsion subjected to different circulations in HPH

As expected, increasing the number of passes through the homogenizer resulted in progressively smaller particles. A single circulation (1x) achieved a significant reduction of 39%, bringing the initial size of 16.106 µm down to 9.796 µm. Further reductions were observed with two and three passes (2x and 3x), reaching final sizes of 8.656 µm and 8.008 µm. It signifies 53% and 56% decrease (Figure 18). This is attributed to the intense shear forces within the homogenizer that disrupt and break down larger droplets into smaller ones.

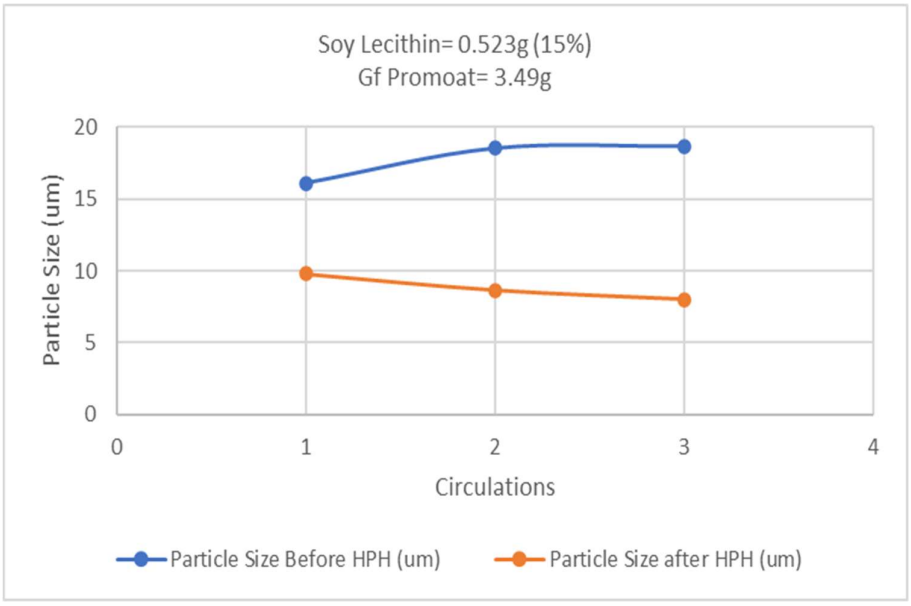

Figure 18; Droplet size of emulsions undergone different circulations in HPH

Conversely, all HPH treatments led to a decrease in apparent viscosity. The initial viscosity of the emulsions ranged from 1.179 to 1.259 Pas. After a single pass, a substantial reduction of 46% was observed, with the viscosity dropping to 0.6827 Pas. Further homogenization resulted in additional

reductions at 2x (0.6602 Pas) and a more significant decrease of 61% at 3x (0.4908 Pas) (Figure 19). This decrease in viscosity can be explained by the formation of smaller and more uniformly sized droplets, offering less resistance to flow. However, the impact on viscosity seemed to diminish with additional passes, suggesting a potential point of diminishing returns in terms of processing time and energy efficiency.

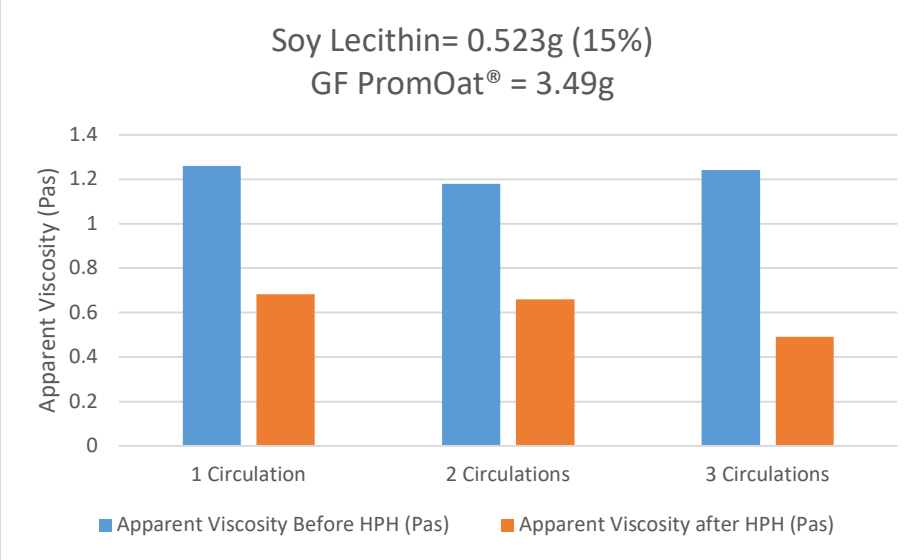

Figure 19; Apparent viscosity of emulsions with varying number of circulations during HPH

Importantly, visual observation indicated that all emulsions-maintained stability for at least two weeks following HPH treatment, irrespective of the number of circulations employed as shown in the figure 20. This suggests that all homogenization conditions achieved sufficient particle size reduction to promote stability during the observation period. This stability is crucial for the long-term shelf life and functionality of the emulsions in various applications.

Figure 20; Emulsions with different number of circulations

4.4 Assessing Emulsion Stability

4.4.1 Variation of Viscosity due to HPH conditions

The viscosity of the oat β-glucan emulsions significantly decreased after high-pressure homogenization (HPH) treatment compared to the unhomogenized state. As shown in Table 15. The apparent viscosity of Emulsion A (1.187 Pas) decreased with increasing HPH pressure (100 bar, 300 bar, and 700 bar).

Emulsion	Pressure (bar)	Apparent Viscosity Before Homogenization (Pas)	Apparent Viscosity after Homogenization (Pas)
Emulsion A	100	1.187	0.4988
Emulsion A	300	1.187	0.4356
Emulsion A	700	1.187	0.2197

Table 15; The apparent viscosity (η) of oat β-glucan-containing emulsions at 100, 300, and 700 bar before and after HPH.

HPH treatment disrupts the network formed by β-glucan particles and reduces the size of oil droplets within the emulsion (Kivelä et al., 2010). This disruption can be explained by two key mechanisms. During HPH, the emulsion is subjected to intense shear forces caused by the high pressure and the design of the homogenizer valve. These forces act to break down the β-glucan network, reducing the entanglement and interaction between β-glucan

molecules. This weakening of the network structure allows for easier flow and hence, a decrease in viscosity (Kivelä et al., 2010).

The rapid pressure fluctuations within the HPH chamber can lead to cavitation, a phenomenon where bubbles form, grow, and collapse violently. The collapse of these bubbles generates intense shockwaves that can further disrupt β-glucan aggregates and contribute to the reduction of oil droplet size. Smaller and more uniform oil droplets offer less resistance to flow within the emulsion, further contributing to the decrease in viscosity. The results support these mechanisms. The progressive decrease in viscosity with increasing HPH pressure (greatest reduction at 700 bar) suggests a stronger disruption of the β-glucan network and more significant reduction in oil droplet size at higher pressures.

The ability to control viscosity through HPH is crucial for various applications in the food industry. Oat β-glucan emulsions are increasingly used in functional beverages. HPH allows for tailoring the viscosity to achieve a desirable mouthfeel, ranging from thin and refreshing to thick and creamy. Viscosity plays a role in various functionalities of foods. HPH can be used to design functional foods with specific rheological properties, such as controlled spreadability in spreads or improved stability in yogurt formulations containing β-glucan.

4.4.2 Determination of the Stability

Three distinct sets of emulsions were formulated using identical concentrations of GF PromOat® combined with soy lecithin, serving as the emulsifying agent, as outlined in the table 6. To assess the migration phenomena, a macroscopic characterization was conducted on all samples using the Turbiscan® LAB Analyzer. An assessment of these emulsions' stability under HPH at pressures of 100 bar, 300 bar, and 700 bar is given in the discussion that follows.

Stability Analysis of emulsion treated with 100 bar pressure

The analysis of the emulsion treated with HPH at 100 bar revealed a clear case of creaming. During the initial scans (represented by the blue curve) (figure 21), the backscattering signal was uniform throughout the sample. This indicates a well-dispersed emulsion, where the oil droplets are evenly distributed within the continuous aqueous phase. However, subsequent scans (From green to red curve) displayed a significant change. The backscattering intensity decreased at the bottom of the cell, while a corresponding increase

was observed at the top. This distinct pattern signifies the migration of oil droplets upwards – a phenomenon known as creaming.

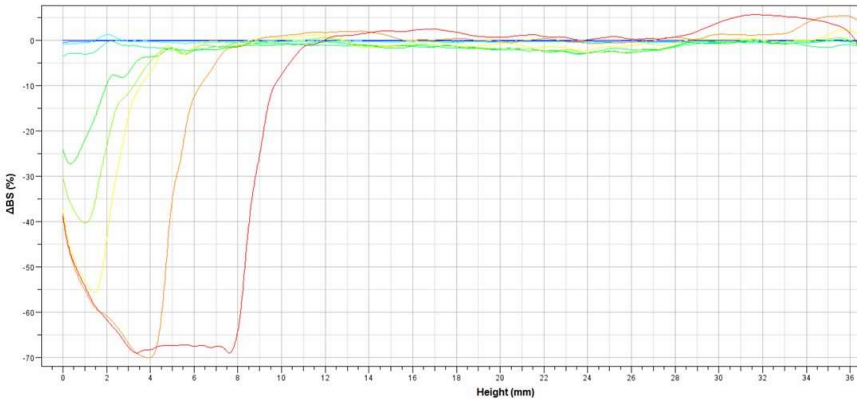

Figure 21; Sample from the Turbiscan Lab Analyzer treated at 100 bar, analyzed in a single scan.

The Turbiscan analysis, particularly the peak thickness function offers valuable insights into the creaming behavior of the 100 bar emulsion (figure 22). A constant peak thickness observed for the initial two days signifies a state of good stability. This indicates that the oil droplets remained well-dispersed throughout the aqueous phase during this period.

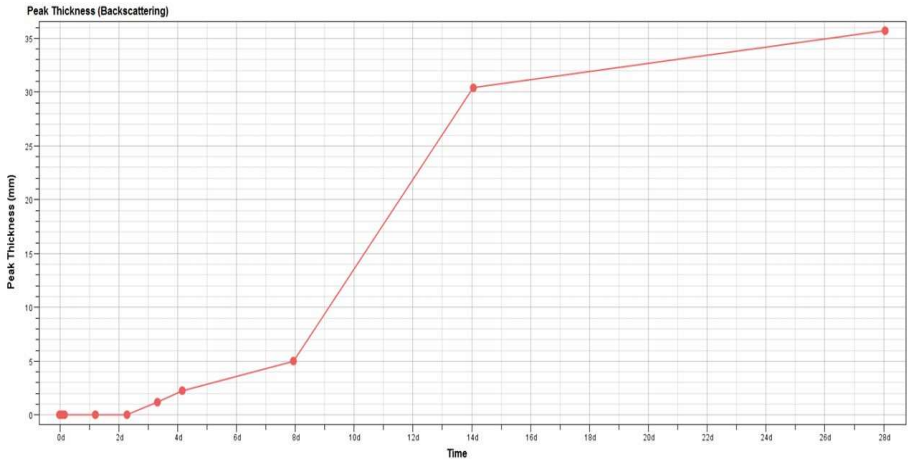

Figure 22; Peak thickness as obtained from Turbiscan Lab analyzer

However, after this initial stable period the peak thickness function begins to increase. This change signifies the onset of creaming. As in creaming the oil droplets, being less dense than the surrounding water phase, migrate upwards due to buoyancy forces. The increasing peak thickness reflects this migration. The function measures the height difference between the bottom of the sample and the point with the highest concentration of oil droplets, as detected by backscattering intensity. As oil droplets accumulate towards the top of the emulsion, the zone with the maximum backscattering intensity also moves upwards, resulting in a larger peak thickness value.

By the eighth day, the peak thickness reaches 5 mm, suggesting a substantial accumulation of oil droplets at the top of the sample. This significant value indicates a clear separation between the oil and aqueous phases. Finally, the complete separation observed during the second week confirms the total destabilization of the emulsion. The oil phase formed a distinct layer on top of the aqueous phase, with minimal or no oil droplets remaining dispersed throughout the bottom portion (figure 23). Since oil is typically less dense than water, the oil droplets experience a buoyant force that pushes them upwards. This phenomenon is well documented in the field of food emulsions, where maintaining stability is crucial for product quality and shelf life.

Figure 23; Emulsion showing Separation after 30 days

The observed creaming behavior in the 100 bar treatment can be attributed to the relatively large size of the oil droplets (12.044 μm)(Table 16). Studies have shown a clear link between droplet size and creaming susceptibility. Emulsions containing larger droplets like those in the 100 bar treatment, experience greater creaming compared to emulsions with smaller droplets.

Pressure (Bar)	Droplet Size Before Homogenization (Vol. Weighted Mean D[4,3])(µm)	Droplet Size after Homogenization (Vol. Weighted Mean D[4,3])(µm)
100	16.106	12.044
300	16.106	8.656
700	16.106	4.035

Table 16; Droplet size of samples before and after HPH at different pressures

Stability Analysis of emulsion treated with 300 bar pressure

The HPH treatment at 300 bar effectively reduced the oil droplet size in the emulsion from a relatively large 16.106 µm to a significantly smaller 8.656 µm (table 15). This notable decrease in droplet size is crucial for enhancing stability due to several key factors. Firstly, smaller droplets experience a weaker buoyant force because of their lower mass. It delays their migration upwards through the aqueous phase thus improving overall stability (McClements, 2015). Secondly, the reduction in droplet size increases the interfacial area between oil and water phases. It facilitates better interaction with emulsifiers and potentially enhancing emulsion stability (McClements, 2017).

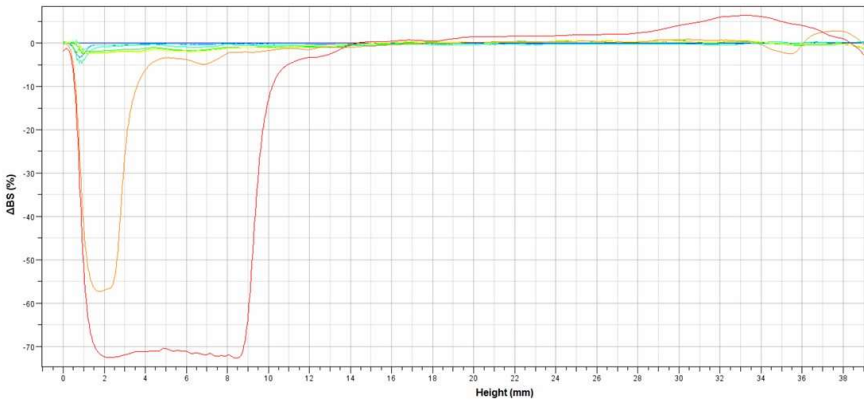

Figure 24; Sample from the Turbiscan Lab Analyzer treated at 300 bar, analyzed in a single scan.

Despite the promising reduction in droplet size, the stability was seen for only the initial two weeks. The stability is apparent from green and blue curves and after that the sample creamed (orange and red curve) (figure 24). This indicates the presence of other influencing factors contributing to creaming.

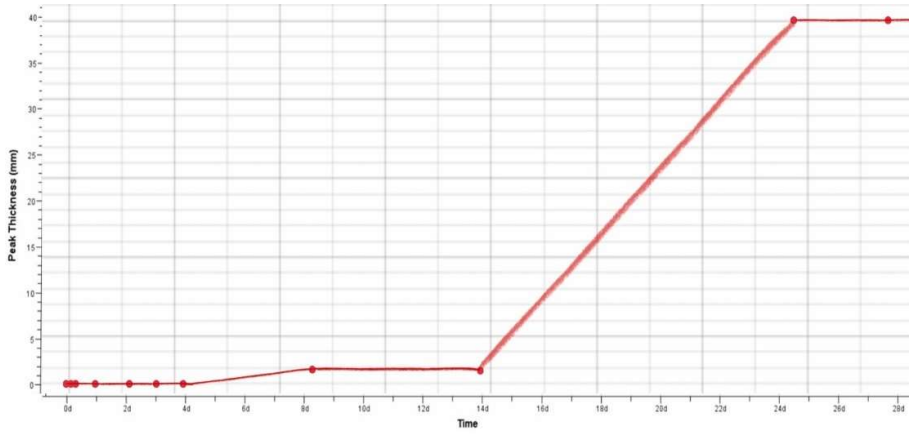

Figure 25; Peak thickness as obtained from Turbiscan Lab analyzer for 300 bar.

The peak thickness analysis sheds light on the creaming behavior of the 300-bar emulsion (figure 25). A constant peak thickness for the initial 14 days signifies a state of good stability, with oil droplets remaining well-dispersed throughout the aqueous phase. However, after this initial stable period the peak thickness starts to increase. This indicates the onset of creaming, where oil droplets begin to migrate upwards due to their buoyancy. The increasing peak thickness reflects this movement as the function measures the height difference between the bottom and the zone with the highest oil droplet concentration. By the third week, complete creaming was observed. This signifies a rapid acceleration of the creaming process and total destabilization of the emulsion. There was a significant accumulation of oil droplets at the top and a clear separation between the oil and aqueous phases.

The delayed creaming onset (after 14 days) in the 300-bar treatment compared to the 100-bar treatment can be attributed to two main factors. Firstly, the 300-bar treatment achieved a smaller droplet size (8.656 μm) compared to the 100-bar treatment. Research found that the smaller droplets experience a weaker buoyant force, delaying their migration and creaming initiation (McClements, 2015). Secondly, the smaller droplet size might allow the soy lecithin (emulsifier) to provide more effective steric hindrance

initially. This could create a better barrier around the droplets, temporarily preventing aggregation and delaying creaming.

Despite the initial stability, creaming eventually occurred due to potential limitations (figure 26). Over time, the Soy lecithin molecules adsorbed onto the droplet surface might become depleted or detach due to various interactions within the emulsion. This depletion or detachment could reduce the emulsifier's effectiveness and lead to creaming (McClements (2017). Additionally, the initial 14-day stability period could represent a metastable state. Even with a reduced size, the smaller droplets might require some time to overcome energy barriers before initiating a more rapid creaming process.

Figure 26; Emulsion showing Separation after 30 days

Stability Analysis of emulsion treated with 700 bar pressure

The application HPH at 700 bar resulted in a substantial reduction of oil droplet size within the emulsion, diminishing them to a mere 4.035 μm (table 15). This pronounced reduction played a pivotal role in sustaining the emulsion's stability over a prolonged observation period of 30 days (figure 27). The rationale underlying this phenomenon is straightforward. Smaller droplets exhibit reduced buoyant forces. This mitigates their propensity to ascend through the aqueous phase, consequently minimizing creaming.

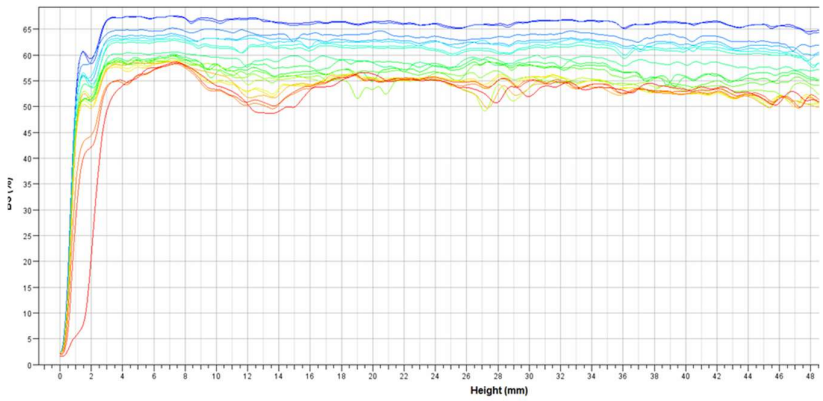

Figure 27; Sample from the Turbiscan Lab Analyzer treated at 700 bar, analyzed in a single scan.

The 700-bar treatment achieved a droplet size small enough to overcome several factors that could lead to instability. Firstly, the smaller droplets allow the Soy lecithin to provide more effective coverage. This improved steric hindrance prevents the droplets from aggregating and destabilizing the emulsion. Secondly, the reduced size lowers the collision rate between droplets due to their decreased mobility within the system. This minimizes the chance of droplet aggregation, further enhancing the kinetic stability of the emulsion.

Figure 28; Emulsion showing stability after 30 days

The combination of a minimized buoyant force and improved steric hindrance due to smaller droplets contributed to the emulsion's stability. Additionally, enhanced kinetic stability played a role. These factors led to the remarkable long-term stability observed in the emulsion treated with 700 bar pressure (Figure 28)

4.5 Packaging Evaluation of GF PromOat®

4.5.1 Packaging, Labelling and claims evaluation

Figure 29; GF PromOat® Packaging

GF PromOat® packaging is designed with specific dimensions of 520 x 880 x 140 mm. It is constructed as a 4-ply open mouth sack, with one of the plies being a plastic liner on the inside. The outer structure of the sack consists of 3-ply unbleached kraft paper, each ply having a weight of 70 g/m². The overall tear energy absorption (TEA) of the sack averages 584 J/m², ensuring durability and resistance to wear and tear. The internal plastic liner is made of 100 µm blue-tinted low-density polyethylene (LDPE), featuring a welded bottom to enhance its strength and prevent leaks.

The 4-ply packaging is renowned for its ability to provide comprehensive containment, protection, and functionality across various products. Its robustness stems from the LDPE layer, serving as an effective shield against moisture, oxygen, light, and pests. LDPE stands out as the preferred choice for moisture-sensitive or oxygen-sensitive items such as food powders or pharmaceuticals. Different studies, like the one conducted by Wong et al. (2020), underscore efficacy in maintaining product stability, as evidenced by the successful preservation of honey jackfruit powder. However, it's imperative to achieve a proper seal between the paper and plastic layers to optimize barrier properties. As noted by Robertson (2006), any imperfections in seals can compromise the overall protective capabilities of LDPE packaging.

Containment

The pouch packaging used by GF PromOat®, offers a compelling solution for containing the oat beta-glucan powder. This material boasts excellent barrier properties against moisture, oxygen, and light (Wong et al 2020). These properties are crucial for preserving the oat beta-glucan. Moisture exposure can lead to undesirable clumping and microbial growth, while oxygen can degrade the product itself. Light exposure might also diminish some of the health benefits associated with oat beta-glucan (Chen et al., 2018). By acting as a strong barrier, the 4-ply packaging directly contributes to extending the shelf life of the oat beta-glucan powder.

However, there are some potential weaknesses to consider with this packaging. Punctures or tears in the pouch can compromise containment, allowing moisture, oxygen, or even contaminants to enter. Additionally, the quality of the seals between the plastic and paper layers is critical. Imperfect seals can create pathways for these same elements to bypass the barrier.

Protection

The 4-ply material used in GF PromOat® 's packaging offers a significant benefit in terms of light protection. Research highlights the effectiveness of this material in shielding products from light degradation (Marsh and Bugusu, 2007). This is particularly important for oat beta-glucan powder, as some studies suggest that light exposure might diminish its health benefits (Chen et al., 2018). By minimizing light exposure, the packaging helps to preserve the potential health-promoting properties of the oat beta-glucan.

However, it's important to acknowledge a weakness of pouch packaging when it comes to physical protection. Compared to rigid containers, pouches like the one used by GF PromOat® might offer limited defense against external impacts or crushing. This could be a concern if the product is susceptible to damage during transportation or handling. If physical protection from external factors is a major concern for GF PromOat®, they could consider additional packaging solutions. Including the pouch within a cardboard box for secondary packaging would create a more robust barrier against physical damage. This approach would balance the excellent light protection offered by the ALP pouch with the enhanced physical protection provided by a rigid container.

Apportionment

While the pouch format used by GF PromOat® offers some advantages for portion control, there's room for improvement. Gf PromOat pouches can be

pre-formed in various sizes, allowing for the creation of single-serving units, as evidenced by research on sliced apples packaged in similar pouches (Bhatt, 2020). This eliminates measuring for consumers and ensures consistent dosing. Additionally, pouches are compatible with including a measuring scoop, which can be a reusable tool for accurate portion control, especially if the pouch itself isn't pre-divided.

The resealable feature makes these units much more practical. Consumers can enjoy single doses and then reclose the pouch for later use, eliminating concerns about accurately measuring or maintaining freshness for the remaining powder. Additionally, the resealable format still allows for including a measuring scoop within the packaging. This reusable tool provides a convenient and mess-free way to accurately measure out desired serving sizes, even if the pouch itself isn't pre-divided. While pre-measured compartments within the pouch could be an option for single-serve convenience, the resealable pouch already offers a significant improvement for portion control. Including a measuring scoop alongside the resealable pouch would be the most impactful way for GF PromOat® to further enhance user experience.

4.5.2 Labelling and Claims

Figure 30; Lowers cholesterol claim on the GF PromOat® 's packaging

With the rise in obesity, heart disease, and diabetes mellitus, consumers are increasingly prioritizing a healthy lifestyle, emphasizing a balanced diet over medication. This shift has spurred innovation in the food industry, particularly in delivering health benefits through food and beverages. Beta-glucan, a soluble fiber found in oats, oat bran, barley, and barley bran, plays a crucial role. Scientifically proven to lower cholesterol, beta-glucan achieves this by absorbing bile acids and salts in the intestines, facilitating

their removal from the body. The European Food Safety Authority (EFSA) and the US Food and Drug Administration (FDA) have granted health claims for beta-glucan due to its cholesterol-lowering effect (figure 30).

According to EU Regulation, in order for a food product to make this claim, it must contain at least 1 g of beta-glucans from the mentioned sources per quantified portion. Additionally, to convey the beneficial effect, the consumer must be informed that a daily intake of 3 g of beta-glucans from these sources is necessary. (EFSA Journal numbers 2009; 7(9):1254 and 2011;9(6):2207).

Another health claim permitted by EU regulation regarding beta-glucans from oats and barley pertains to the reduction of the blood glucose rise after a meal. For a food product to make this claim, it must contain at least 4 g of beta-glucans from oats or barley for each 30 g of available carbohydrates in a quantified portion as part of the meal. Similarly, the consumer must be informed that the beneficial effect is obtained by consuming the beta-glucans from oats or barley as part of the meal (EFSA Journal 2011;9(6):2207)

The packaging could use the marketing terms emphasizing the fact that Innovations like Gluten-Free PromOat® cater to consumers with gluten restrictions, ensuring they can still enjoy beta-glucan's health benefits. Besides cholesterol reduction and glycemic control, beta-glucan supports digestive health by promoting the production of butyric acid during colonic fermentation, essential for colon cell health and function. Studies also suggest that beta-glucan-rich foods and drinks may enhance satiety, aiding weight management efforts. With obesity rates on the rise, incorporating beta-glucan into various products offers a promising solution to address this pressing public health concern.

Gluten Free Claim

In compliance with EU regulations, GF PromOat® proudly bears the "gluten-free" label, adhering to the stringent standards outlined in Regulation (EU) No 828/2014. This regulation stipulates that for a product to be designated as "gluten-free," it must contain less than 20 parts per million (ppm) of gluten. This threshold is established to safeguard individuals with gluten sensitivities or celiac disease, ensuring that products labeled as gluten-free pose minimal risk of adverse reactions. The 20-ppm limit is widely recognized as a safe level of gluten exposure for most individuals with celiac disease, as

supported by scientific research and consensus within the medical community.

GF PromOat®'s commitment to compliance with these regulations is unwavering. Rigorous testing protocols are implemented to verify the gluten-free status of every batch of GF PromOat®. These protocols encompass both raw materials and finished products, utilizing validated analytical methods to accurately measure gluten levels. By meeting the criteria for gluten-free labeling set forth by EU regulations, GF PromOat® provides consumers with a trustworthy option that aligns with their dietary requirements. This assurance is particularly crucial for individuals who must strictly adhere to a gluten-free diet to manage their health conditions effectively.

5 Conclusions, limitations and recommendations

5.1 Conclusion

Developing successful food products requires a deep understanding of how ingredients interact within a complex food system. Food emulsions, due to their intricate nature, often exhibit thermodynamic instability. This is a result of the inherent properties of their components. Therefore, selecting the right emulsifier and thickening/stabilizing agent is crucial to ensure the emulsion's long-term stability, considering both the formulation itself and the processing conditions.

This study explored different combinations and possibilities to make emulsions using 25% of rapeseed oil. 15% of soybean lecithin (based on the amount of GF PromOat® were used at pH 6.2. The aim was to identify the optimal concentration range of GF PromOat® that could be used in these particular conditions.

The critical overlap concentration (c*) of GF PromOat® in solution was determined to be 0.0349 g/mL by examining its rheological behavior. The samples showed steep increase in apparent viscosity upon reaching the c*. However, the stability of the samples was not only dependent on the concentration of GF PromOat®. The appropriate processing conditions and suitable combination with emulsifier were also significant. These conditions included high-pressure homogenization (HPH) at a 700 Bar pressure with three homogenization cycles.

In conclusion, HPH at 700 bar significantly reduced oil droplet size and enhanced stability over 30 days. Smaller droplets experience reduced buoyant forces, which minimizes creaming. The reduced droplet size enhanced soy lecithin coverage, which provided better steric hindrance. It also decreased droplet collision rates, which reduced aggregation. These factors collectively contributed to the emulsion's remarkable long-term stability observed in the 700-bar treatment.

5.2 Limitations and Future Research

Food emulsions are complex systems, with underlying stabilizing and destabilizing mechanisms defined by the interactions between their constituent parts. While this study sheds light on specific emulsion characteristics, it is crucial to approach the results with caution, considering the unique nature of the emulsion under examination.

Moreover, future research should explore a wider range of emulsions to ascertain the generalizability of the findings. Specifically, investigating emulsions with varying oil fractions is essential to understand the hydrodynamic effects caused by the presence of oil droplets. Notably, the emulsions examined in this study contained unusually high amounts of oil, a factor that may not align with typical food applications, especially in the production of plant-based beverages.

Therefore, a comprehensive exploration of emulsions with more typical oil concentrations is warranted to better understand the practical implications of the observed stabilizing and destabilizing mechanisms. This approach will provide valuable insights into the behavior of emulsions in real-world food production scenarios, ensuring the applicability of the findings across a broader spectrum of products.

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Appendix A:

Emulsions containing different amounts of Soybean lecithin relative to the amount of Gf PromOat.

Soy Lecithin	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.35g(20% of 1.75)	1.75	25	72.9
0.7g(40% of 1.75)	1.75	25	72.5
1.05g(60% of 1.75)	1.75	25	72.2
1.4g(80% of 1.75)	1.75	25	71.8
1.75g(100% of 1.75)	1.75	25	71.5

Soy Lecithin	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.131g(5% of 2.62)	2.62	25	72.2
0.262g(10% of 2.62)	2.62	25	72.1
0.524g(20% of 2.62)	2.62	25	71.8
1.048g(40% of 2.62)	2.62	25	71.3
1.572g(60% of 2.62)	2.62	25	70.8
2.096g(80% of 2.62)	2.62	25	70.2
2.62g(100% of 2.62)	2.62	25	69.7

Soy Lecithin	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.1745g(5% of 3.49)	3.49	25	71.3
0.349g(10% of 3.49)	3.49	25	71.1
0.698g(20% of 3.49)	3.49	25	70.8
1.396g(40% of 3.49)	3.49	25	70.1
2.094g(60% of 3.49)	3.49	25	69.4
2.8g(80% of 3.49)	3.49	25	68.7
3.49g(100% of 3.49)	3.49	25	68.0

Soy Lecithin	GF PromOat® (g)	Rapeseed Oil (g)	Buffer pH 6.2 (g)
0.026g(0.5% of 5.25)	5.25	25	69.7
0.0525g(1% of 5.25)	5.25	25	69.5
0.1312g(2.5% of 5.25)	5.25	25	69.4
0.2625g(5% of 5.25)	5.25	25	69.3
0.525g(10% of 5.25)	5.25	25	69.2