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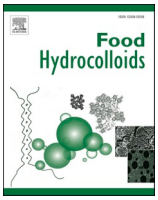

Formulation of high protein pea gels: Oil and microgel particles from pectin modulate the rheological, tribological and optical properties

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ABSTRACT

Pea protein is becoming increasingly important in the food industry due to various factors. The majority of vegan alternatives to dairy products lack the same texture as their conventional counterparts. This issue is particularly evident in the context of vegan yogurt alternatives and high protein alternatives. It is therefore crucial to be able to modify the texture of vegan alternatives, such as pea-based yogurt alternatives with a high protein content.

This study examined the potential for modulating the texture of fermented pea protein gels through the use of canola oil and pectin-based microgel particles (MGP). To achieve this objective, vegan yogurt alternatives with a high protein concentration ($\varphi_{PPI} = 12$ wt%) were produced and their tribological and rheological properties were characterized.

The results showed that adding various concentrations of canola oil improved lubrication and color, but increased dynamic viscosity as well as gel strength. The addition of 1 μm pectin-based microgel particles, which act as inactive fillers, reduced dynamic viscosity without negatively affecting lubrication.

The combination of these two results in enhanced lubrication in pea protein yogurt alternatives, while simultaneously reducing the dynamic viscosity and gel strength. These findings suggest potential strategies for modulating vegan high protein yogurt alternatives through the addition of canola oil and pectin-based microgel particles.

1. Introduction

Yogurt is one of the world's most popular dairy products and contributes to a healthy diet thanks to its high nutrient content (McKinley, 2005). In recent years, the demand for vegan foods has increased for reasons of sustainability, animal welfare, but also dairy intolerance. As a result, attempts are being made to replace more and more animal proteins with plant-based proteins, including those in yogurt. Vegan yogurt alternatives made from soy, coconut or almond proteins are widely available on the market (Dhakal et al., 2023). Pea protein is becoming increasingly important in the food industry, due to its low allergenicity, regional high availability, low cost and health benefits (Lu et al., 2020). However, pea protein as a food ingredient still poses challenges in terms of functionality, taste and color, which have not been solved yet (Lam et al., 2018; Zhao et al., 2020). Therefore, the utilization and characteristics of pea protein as a fundamental component in yogurt alternatives have been largely overlooked until recently. To produce plant-based yogurt alternatives, starter cultures are needed to ferment the plant protein dispersion (Mecray, 2022; Yazici et al., 1997). The bacteria produce lactic acid, which lowers the pH from 4.5 to 5.0 (Shen et al., 2021). In this pH range, the proteins agglomerate due to the reduction of electrostatic repulsion and a protein gel is formed (Tay et al., 2005). The characteristics of the resulting protein gels, including gel strength and lubrication, have a direct impact on the properties of the yogurt alternative. One of the most important quality attributes of consumer acceptance and preference of food products is the oral perception of texture (Surmacka Szczesniak, 2002). Therefore, the production of vegan yogurt alternatives is not only about replacing animal proteins with plant proteins, but also about creating a satisfying texture and expected mouthfeel. Yogurt alternatives often have a texture that is described as too firm or too soft and a mouthfeel that is perceived as sandy and watery (Greis et al., 2023; Yang et al., 2012; Yousseef et al.,

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2016).

The flow properties in the mouth and the associated sensory perception can be described by rheology. However, as soon as the particle diameter of food decreases due to oral processing, friction between mouth surfaces becomes the dominant mechanism of mouthfeel and can no longer be captured by rheological parameters. The lubricating properties of the food are described and interpreted by oral tribological measurements (Kokini et al., 1977). It enables the measurement of the forces between two surfaces in contact during relative motion, as well as the effect of a lubricant between those surfaces (Stokes et al., 2013). The result of a tribological analysis is a Stribeck curve, in which the friction coefficient is plotted against the relative velocity. The Stribeck curve is further categorized into three distinct regimes (from left to right): the boundary, mixed, and hydrodynamic regime. In the boundary regime, high friction coefficients predominate as the two surfaces of the tribo geometry are in contact at slow relative velocity. Conversely, at the extremities of the range, a lubricating film accumulates, thereby reducing the friction coefficient as the velocity increases. At the onset of the mixed regime, a continuous lubricating film is established. As the relative velocity increases further, the hydrodynamic action of the lubricant becomes more dominant, indicating a transition to the hydrodynamic regime. In this final regime, the friction continues to decrease, and the contact surfaces are separated from each other by the formation of a thicker lubricating film. It is notable that at the end of the tribological measurements, an increase in the friction coefficient is observed, attributable to the hydrodynamic behavior of the lubricant. The friction and lubricating properties of food depend on various parameters, such as the addition of fat, particle size or particle deformation (Krzeminski et al., 2012; Liu et al., 2016; Selway & Stokes, 2013; Wijk & Prinz, 2005). With reduced friction, the consumer perceives a creamier mouthfeel (Prakash et al., 2013). The mouthfeel encompasses all tactile properties that are perceived when the food is placed in the mouth and chewed until it is swallowed (Guinard & Mazzucchelli, 1996; Stokes et al., 2013). To make the mouthfeel of a plant-based yogurt alternative creamier and less sandy, fillers like oil or soluble fiber in the form of microgel particles can be added (Saavedra Isusi et al., 2023; Sonne et al., 2014). The particulate structure of microgel particles is expected to allow them to roll between the tongue and palate, thereby improving mouthfeel (Sarkar et al., 2017). Moreover, yogurt with a higher fat concentration has better lubricating properties (Sonne et al., 2014). According to Dickinson's model (Dickinson & Chen, 1999), there are basically two ways in which an added substance can affect the gel network. The first possibility is that the added substance is incorporated into the protein network as a so-called active filler and strengthens the gel structure through strong interactions with it. The other possibility is that the added substance has no chemical affinity to the built-up network and weakens the gel network as an inactive filler (Dickinson & Chen, 1999). If the yogurt alternative is produced with oil, fermentation results in an emulsion-filled gel. This is defined as a complex colloidal system formed by the combination of an emulsion and a gel phase (Dickinson, 2012). In the case of the pea protein-based yogurt alternative, the continuous phase is aqueous. The oil droplets are dispersed in this phase. The pea protein dissolved in the water adsorbs to the oil droplets and forms a viscoelastic layer that acts as a physical barrier to prevent instability phenomena (Wilde et al., 2004). It is assumed that oil acts as an active filler and thus strengthens the gel structure. The pea protein isolate (PPI) around the oil droplets can interact with the pea protein in the continuous phase and thus strengthen the gel structure (Dickinson, 2012). It is known from the literature that the addition of high oil concentrations (20 wt%) to emulsion-filled gels based on soy protein leads to harder gels (Gu et al., 2009). The addition of only coconut oil (4 wt%) to a fermented emulsion gel based on soy protein isolate, however, does not result in a harder gel (Saavedra Isusi et al., 2022). Fibers such as pectin, which is the raw material for the pectin-based microgel particles, can also be used as a thickening agent in its original form (EFSA Panel on Dietetic Products, Nutrition & Allergies, 2010; Pang et al., 2020). Pectin is obtained from side streams of juice and sugar production and is therefore a sustainable fiber. Nevertheless, if added to protein matrices, the polymer strands cannot produce a creaminess comparable to that of fat globules (Pang et al., 2020; Tomic et al., 2017). Kieserling et al. (2019) showed that the addition of particulate dietary orange fibers has no effect on the creaminess of yogurt, although the friction increases (Kieserling et al., 2019). Microgels based on biopolymers have a functional application in food due to their mouldability, surface activity, reversible swelling and reactivity to pH and temperature (Dickinson, 2015). Saavedra Isusi et al. (2023) showed that pectin-based microgel particles can be used in plant-based gels (Saavedra Isusi et al., 2023). In one of their studies, the effect of microgel particles was examined in soy-based vegan yogurt alternatives which had a protein content of 5 wt% (i.e. non-high protein). It was demonstrated that the addition of 3 wt% or 5 wt% pectin-based microgel particles weakened the structure of the soy protein gel because they served as inactive fillers (Saavedra Isusi et al., 2023). At pH values above 2, pectin-based microgel particles are negatively charged due to their free carboxyl groups (Saavedra Isusi et al., 2021). As the isoelectric point (IEP) of the two main fractions of PPI (vicilin at pH 4.8 and legumin at pH 5.5) is within the same range as that of the soy protein isolate fractions (β-conglycinin at pH 4.64 (Koshiyama, 1968), and pH 4.9 for glycinin (Peng et al., 1984). Consequently, it can be deduced that the repulsive interactions observed in SPI matrices will also be evident in PPI matrices (Klost & Drusch, 2019; Klost et al., 2020).

The aim of this study is to investigate how the texture of a vegan high protein yogurt alternative based on PPI can be modulated. For this purpose, MGP, oil and a combination of both are added to the PPI matrix before fermentation. It is assumed that the MGP loosens up the structure of the yogurt alternative and the addition of oil improves the lubricating properties and thus the creaminess. The influence of pectin-based MGP on a high protein yogurt alternative based on pea protein has not yet been investigated. The influence of canola oil on a vegan yogurt alternative and the addition of both at the same time is still unclear. Therefore, the questions are systematically investigated and the effects on the rheological and tribological properties are examined.

2. Materials and methods

2.1. Materials

A low methylester sugar beet pectin (SBP) was donated by Herbstreith and Fox GmbH & Co. KG (Neuenbürg, Germany). According to the supplier, the pectin had a degree of esterification of 39 %, a degree of acetylation of 5 % and a galacturonic acid content of 65 %. Calcium chloride di-hydrate (analytical grade) was purchased from Carl Roth GmbH + Co. KG (Karlsruhe, Germany). PPI (Empro® E 86) was kindly provided by Emsland-Stärke GmbH (Emlichheim, Germany) and consists of 80 % protein. Starter cultures (Lactobacillus bulgaricus and Streptococcus thermophilus) were purchased from Metafood GmbH (Frankfurt, Germany). Sucrose was purchased from Carl Roth GmbH + Co. KG (Karlsruhe, Germany). Canola oil was purchased from Bernhard Schell GmbH (Lichtenau, Germany).

2.2. Preparation of pectin solution

To prepare a 2 wt% pectin solution, 98 g demineralized water was added to 2 g sugar beet pectin. The suspension was stirred with a magnetic stirrer (IKA-Werke GmbH & CO. KG, Staufen, Germany) and heated to 70 °C until the pectin was completely dissolved. The solution was then cooled to room temperature.

2.3. Preparation and characterization of pectin MGP suspensions

The addition of calcium chloride ($CaCl_2$) was used to achieve

gelation. To prepare a 40 mM calcium chloride solution, 0.44 g of $CaCl_2$ was added to an 800 mL beaker and dissolved with 99.56 g of demineralized water. The calcium chloride solution was dispersed with an Ultraturrax T-25 digital dispersing rod (IKA-Werke GmbH & CO. KG, Staufen, Germany) at a relative centrifugal acceleration of 2456 RCF (13 000 rpm) while the pectin solution was added. The pectin solution was then dispersed for 3 min.

A 5 wt% MGP solution was prepared. For this, 70 g of the previously prepared pectin solution and 630 g of demineralized water were mixed. The mixture was stirred until all components were completely dissolved and fed into a high-pressure homogenizer (Microfluidizer® Processor M-110 EH, Microfluidics, Newton, Massachusetts, United States of America). The mixture was homogenized twice at a pressure of 600 bar and subsequently filled into bottles and stored in a refrigerator.

The particle size distribution of the MGP was determined by static laser diffraction using a HORIBA LA-950 particle analyzer (Retsch Technology, Haan, Germany) equipped with a flow cell. The scattered light was measured by detectors and the particle size distribution was calculated using Mie theory. Measurements were made in water with a refractive index of 1.547 for the MGP. The particle size distribution was determined in triplicate at room temperature.

2.4. Preparation and characterization of oil in water emulsions

In order to produce the yogurt alternative with oil at a later stage, an emulsion of PPI, canola oil, and demineralized water was prepared in advance. A distinction was made between two emulsion compositions. Emulsion one consisted of 5 wt% PPI, 5 wt% canola oil and demineralized water. Emulsion two consisted of 5 wt% PPI, 11.7 wt% canola oil and demineralized water. The given quantities of PPI and demineralized water were weighed into a 600 mL beaker and stirred. The solution was then dispersed with the high-shear mixer (IKA-Werke GmbH & CO. KG, Staufen, Germany) at a relative centrifugal acceleration of 3270 RCF (15 000 rpm) for 30 s, during which time the canola oil was added and pre-emulsified again for 30 s at 3270 RCF (15 000 rpm). This emulsion was then homogenized at a pressure of 600 bar in a high-pressure homogenizer.

The droplet size distribution of the canola oil emulsion was determined by static laser diffraction using a HORIBA LA-950 Particle Analyzer (Retsch Technology, Haan, Germany), equipped with a flow-through measuring cell. The scattered light was measured by detectors and the particle size distribution was calculated using Mie theory. Measurements were made in water with a refractive index of 1.450 for the canola oil emulsion. The droplet size distribution was determined in triplicate at room temperature. The emulsion containing 5 wt% oil was utilized for the yogurt alternative, which contains 1.5 wt% oil at the end, and the emulsion with 11.7 wt% oil was utilized for the yogurt alternative, which contains 3.5 wt% oil in the finished alternative.

2.5. Preparation of pea protein yogurt alternative

The pea protein gels were produced by fermentation. All pea protein yogurt alternatives ended up containing 12 wt% PPI. For the preparation, PPI and demineralized water were stirred using a magnetic stirrer (IKA-Werke GmbH & CO. KG, Staufen, Germany) with an integrated heat plate until 60 °C was reached. The temperature was held for 30 min. Afterward, the temperature was increased to 80 °C and held for 1 min. If an emulsion was added, this was done before the solution was heated. In this case, 1.5 or 3.5 wt% canola oil was added as an emulsion. The PPI solutions were then cooled down to 45 °C in an ice-water bath. A predetermined amount of MGP solution (0, 1, 2 or 3 wt%) was then added and stirred in. In addition, the yogurt cultures are required for fermentation and 1 wt% sucrose added as a substrate. The solution was then bottled and fermented in a water bath at 43.5 °C for 19 h. After fermentation, the yogurt alternative was stirred smooth at 233 RCF (4000 rpm) for 45 s with the dispersing rod (IKA-Werke GmbH & CO. KG, Staufen, Germany) and placed in the refrigerator for at least 24 h. This allows the gel network to relax, recover and regenerate. All formulations were prepared in triplicate. An overview of the respective compositions can be found in Table 1.

2.6. L*a*b* color space measurements

The color measurements were made with a spectrophotometer (CM-600d, Konica Minolta, Munich, Germany) and evaluated with the CIE L*a*b* system. The color was determined three times.

2.7. pH-measurements

The pH value of the yogurt alternative and the PPI solutions was measured at a temperature between 16 and 19 °C using a pH meter (HANNAH Instruments edge HI2020-02, Vöhringen, Germany) in triplicate.

2.8. Rheological measurements

The rheological properties of the yogurt alternative were analyzed following a 24-h storage period at 5 °C. To obtain information about the structure of the yogurt alternatives, flow curves were measured, and amplitude tests were carried out using a Modular Compact Rheometer (Physica MCR Rheometer 301, Anton Paar, Graz, Austria). For the measurements, a plate-plate geometry with a diameter of 25 mm was used and a gap width of 1 mm was set. Measurements were taken at 20 °C. After removing the yogurt alternative from the refrigerator and putting the sample into the rheometer, a waiting time of 5 min was observed before the first measurement. To determine the dynamic viscosity η a shear stress τ ramp of 0.1 Pa–300 Pa was set. The shear stress τ was increased in steps with a logarithmic ramp of 10 points per decade. A total of 36 measuring points with a measuring point duration of 10 s were recorded. The measurement was carried out three times. The amplitude tests were carried out in duplicate with a constant angular frequency ω of 1 $rad \cdot s^{-1}$ and a shear stress range τ between 0.5 Pa and 100 Pa. The shear stress τ was increased in steps with a logarithmic ramp. The gradient was 10 points per decade. The number of measuring points was 24.

2.9. Tribological measurements

The tribological properties of the yogurt alternative were analyzed following a 24-h storage period at 5 °C. A rotor-stator measuring system consisting of three polydimethylsiloxane (PDMS) pins and a rotating ball was used for the tribological measurements. Specifically, it was a ball-on-3 pins TRI13 45° tribology measurements geometry. The PDMS pins used were manufactured in-house using SYLGRAD TM 184 Silicone Elastomer Kit (Dow Europe GmbH C/O Dow Silicones Deutschland GmbH, Wiesbaden, Germany). To do this, 20 g of SYLGARD Elastomer Base was weighed into a weighing dish and then 2 g of the curing agent was added. It was important to maintain the ratio of elastomer base to

Table 1
Composition of the various yogurt alternatives.

	PPI / wt%	Water / wt%	Sucrose / wt%	MGP / wt%	Canola oil / wt%
Without MGP and oil	12	87	1	0	0
With MGP and without oil	12	86	1	1	0
	12	85	1	2	0
	12	84	1	3	0
Without MGP and with oil	12	85.5	1	0	1.5
	12	83.5	1	0	3.5
With MGP and oil	12	82.5	1	3	1.5

curing agent at 10:1. The two components were stirred for 3 minutes and then degassed in a vacuum oven to avoid air inclusions in the pins. The mixture was then poured into a Teflon mold and the surface was smoothed with a spatula. Care must be taken to ensure that no new air pockets are formed. The PDMS mass in the Teflon mold was cured for 1 h at 100 °C and removed from the mold with a punch after cooling. For measurements, a stress-controlled HAAKE Mars Rheometer (Thermo Scientific GmbH, Karlsruhe, Germany) was used. A glass sphere with a diameter of 125 mm was used as the rotor and the stator was the holder with three PDMS pins. Measurements were taken at a temperature of 20 °C with a rotation speed of 0.0001 mm·s^{-1} to 1000.0 mm·s^{-1} (data shown: conditions in the oral cavity: 10–100 mm·s^{-1}) with a linear, continuous distribution. The measurement duration was 180 s with 1000 data points. The integration time for each data point was 2.0 s. The specified normal force was 1 N. A measurement sequence contained five individual measurements, whereby the first two measurements were seen as running-in for the system and only measurements three, four and five were used for the evaluation. Three measurement runs were made of each yogurt alternative.

2.10. Statistical analysis

Every sample preparation was made in triplicate. If not specified otherwise, all analyses were conducted at least three times per independent test. All data were assessed via multifactorial analysis of variance (ANOVA) and a Scheffé test as a post hoc test. Dissimilarities in the samples were considered statistically relevant at a level of $p \leq 0.05$. The software OriginPro 2020 (OriginLab Corp., Northampton, Massachusetts, United States of America) was used for the statistical analysis, calculation of averages, and standard deviations.

3. Results and discussion

3.1. Size distributions of the microgel particles and the canola oil emulsion

In order to describe the influence of the addition of MGP and/or oil addition, it is important to characterize the MGP and the canola oil emulsion as a first step. The particle size distribution of the pectin-based MGP used is shown in Fig. 1 A. In Fig. 1 B the droplet size distributions of the canola oil-in-water emulsions are shown.

All three curves are bimodal but narrow. The $x_{50,3}$ were found to be 0.97 ± 0.04 µm for the pectin-based MGP, 0.28 ± 0.01 µm and 0.32 ± 0.00 µm for the emulsions 5 wt% and 11.7 wt% oil, respectively. Only minor discrepancies are observed in the large oil droplet fractions. These can be regarded as negligible in subsequent analyses, given that they affect only a minor proportion of droplets.

3.2. Influence of microgel particles or oil addition on the rheological properties of the fermented pea protein gel

In Fig. 2, the dynamic viscosity is plotted as a function of the shear stress for different yogurt alternatives with and without the addition of MGP. All samples exhibited shear thinning behavior and yield stress. Determined was the yield stress at the point at which the plateau value began to decline. The yield stress of the vegan yogurt alternative without the addition of microgel particles is approximately 2.5 Pa (empty squares). With further increasing MGP concentration, the yield stress continues to decline. Ultimately, it is reduced by approximately one order of magnitude to 0.25 Pa with the addition of 3 wt% MGP (filled squares).

A possible explanation is that beet pectin is negatively charged at pH 5. The net surface charge is around 0 at the isoelectric point, which for PPI is at about pH 5 (Danielsson, 1950). The electrostatic interactions

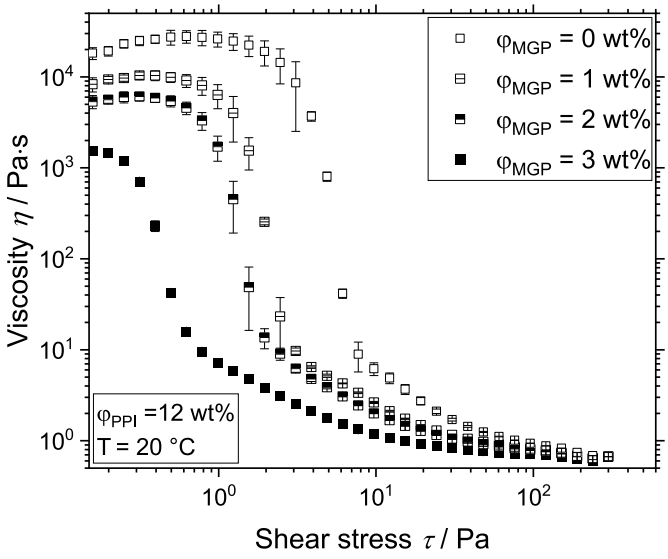

Fig. 2. Flow curves of yogurt alternatives produced without the addition of microgel particles and with 1 wt%, 2 wt% and 3 wt% microgel particles, measured at 20 °C.

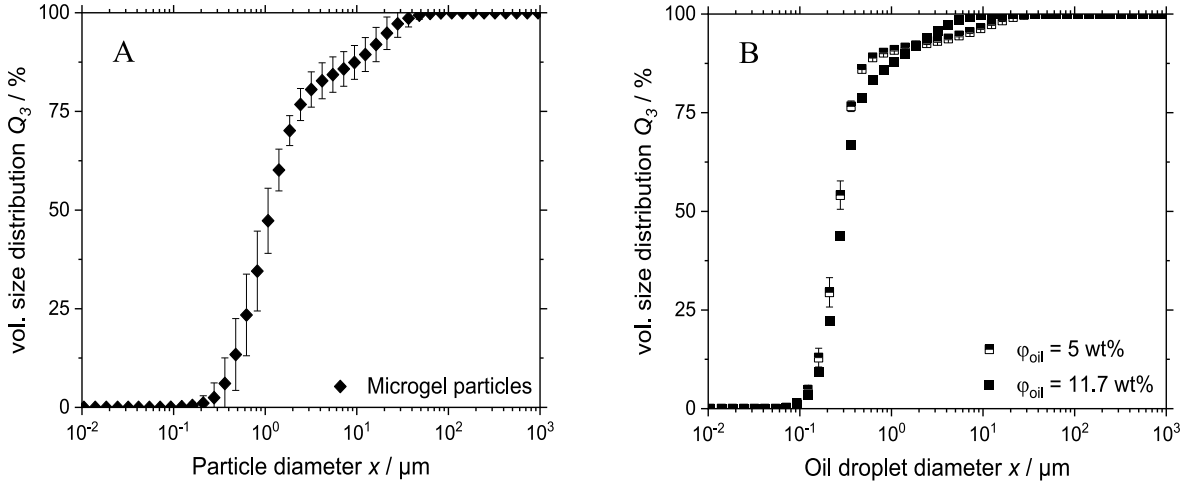

Fig. 1. Volumetric particle/droplet size distribution of (**A**) pectin-based MGP, homogenized at 600 bar and (**B**) canola oil-in-water emulsions with varying oil concentrations and stabilized with PPI.

are therefore limited and the MGP act as inactive fillers in the gel network. Charged particles also repel each other, which can hinder protein aggregation. The greater the number of charged particles in the pea protein suspension, the more obstacles there are to the formation of the protein matrix. In contrast to the results in Fig. 2, Saavedra Isusi et al. (2022) showed for a low concentrate (5 wt%) soy protein gel, that the addition of 1 wt% MGP does not affect the gel network (Saavedra Isusi et al., 2022). This difference may be due to the fact that pea protein forms softer gels compared to soy protein, therefore imperfections in the form of MGP are more noticeable (Shand et al., 2007; Sun & Arntfield, 2010; Tiong et al., 2024). In addition to determining the dynamic viscosity, amplitude tests were conducted to obtain information about the linear viscoelastic (LVE) region. The results of the amplitude tests are presented in Fig. 3. In the LVE, G' > G″ and the sample shows gel-like behavior. At the end of the LVE, G′ decreases and the crossover point (COP) (G' = G″) is created, which is used to determine the point at which the viscous properties of the sample dominate. The length of the LVE region and the maximum shear stress in this region can be used as a measure of the deformability of a gel. The more deformable the gel, the higher the shear stresses that can be absorbed without destroying the sample structure. The addition of MGP leads to a change in the elasticity of the PPI matrix, which indicates a change in the degree of cross-linking (Treloar, 1975).

At low strain stresses, all samples show gel-like behavior which show predominantly elastic behavior. That is, the storage modulus is greater than the loss modulus in all cases. Fig. 3 illustrates that the LVE was shortened and the crossover point was shifted to lower strain stresses by the addition of MGP with stronger effects observable at higher MGP concentrations. Again, in contrast to Saavedra Isusi, the addition of 1 wt % MGP has resulted in a shift of the crossover point, in this case, from a strain stress of ≈ 5 Pa in the absence of MGP (empty symbols) to ≈ 1.7 Pa (empty symbols with horizontal bar). Furthermore, moduli G′ and G″ are reduced with increasing MGP concentration, indicating that the crossover points were reduced. This corresponds to the behavior seen in Fig. 2, where the yield stress shifts to lower shear stresses at higher MGP concentrations.

Figs. 2 and 3 show that the addition of MGP resulted in a significant weakening of the gel network even at low MGP concentrations. MGP act as inactive fillers in pulse protein gels with higher MGP concentrations, leading to increased weakening effects (Saavedra Isusi et al., 2023). However, in contrast to soy gels, pea protein gels seem more sensitive to the presence of MGP. This might be caused by the differences in the gelation mechanism between globular soy proteins and pea proteins, particularly with regard to their distinct functionalities, protein conformations, and compositions (Shand et al., 2007). In comparison with SPI, PPI has a lower protein content due to the presence of more non-protein components, such as starch or fiber (Tiong et al., 2024). Consequently, soy protein has been observed to form stronger gels in comparison to pea protein (Shand et al., 2007; Sun & Arntfield, 2010). A possible explanation could be that the glycinin from soya beans can form a better network than legumin from peas, due to the availability of lysyl and glutaminyl residues (O'Kane et al., 2004).

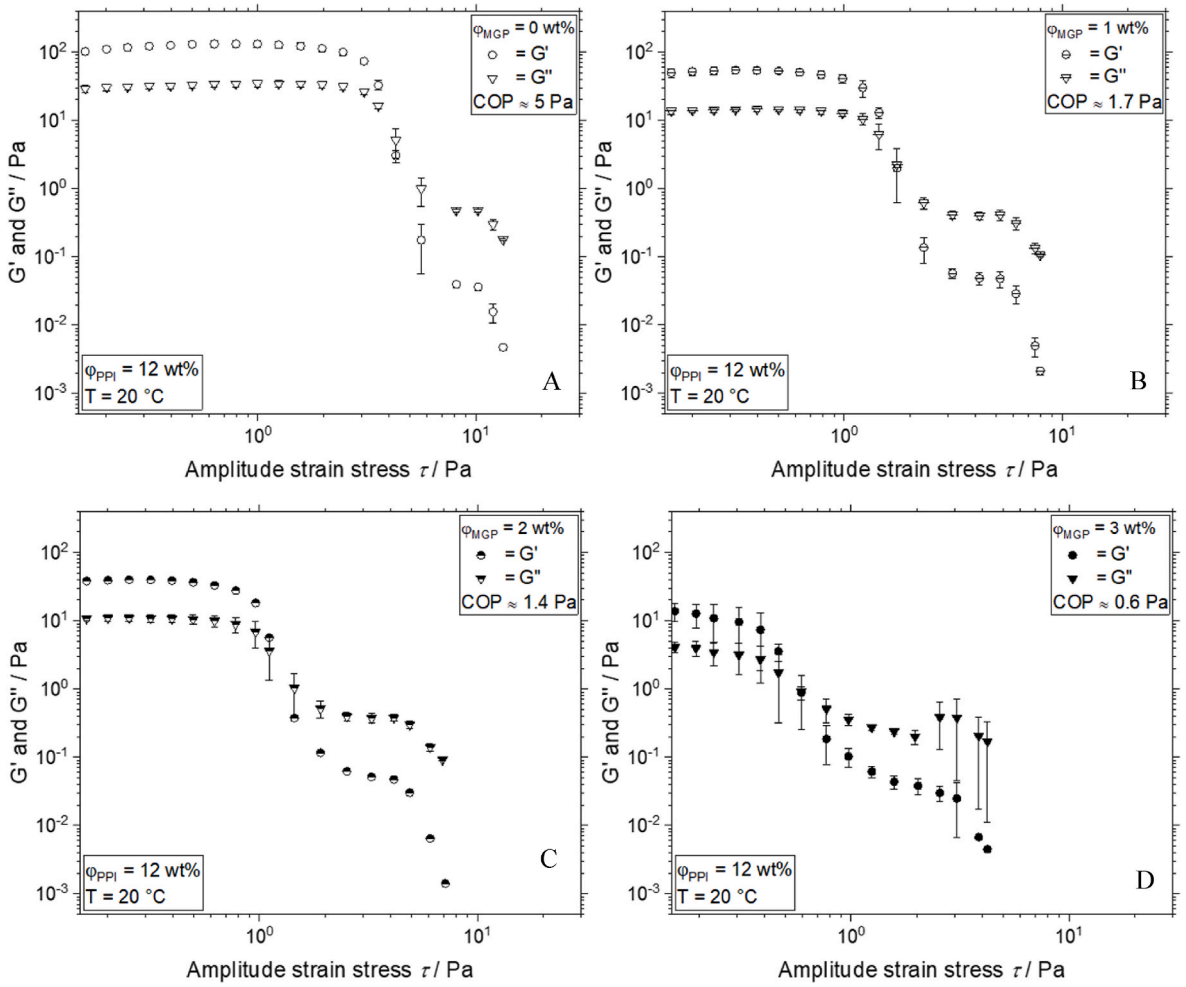

Fig. 3. Amplitude tests of yogurt alternatives produced without the addition of microgel particles (A) and with 1 wt% (B), 2 wt% (C) and 3 wt% MGP (D), measured at 20 °C with a circular frequency of 1 rad·s^{-1}. The values of the crossover points (COP) can be found below the point labels in the legend to the right.

According to Dickinson and Chen (1999), oil is incorporated into a whey protein gel network as an active filler (Dickinson & Chen, 1999). To test whether these observations also apply to the pea protein-based gels, yogurt alternatives with 1.5 wt% and 3.5 wt% oil were prepared and compared with the yogurt alternatives without added oil. It was expected that with increasing oil concentration, the dynamic viscosity increases and the gel network becomes stronger. In Fig. 4, the dynamic viscosity is plotted as a function of the shear stress for vegan yogurt alternatives with and without added oil.

It can be seen, as expected, that with increasing oil content in the yogurt alternative, the yield point increases. Thus, the oil acts as an active filler. An increase from ≈ 1.9 Pa with no oil added (empty squares), to ≈ 3.8 Pa with 1.5 wt% oil (half-filled squares), to ≈ 7.7 Pa with 3.5 wt% oil (filled squares) can be found. This roughly corresponds to a doubling of the yield stress value upon doubling the oil concentration. Dynamic viscosity also increases with increasing oil content. At a shear stress of 40 Pa, the dynamic viscosity is ≈ 1.4 Pa·s with no oil added, compared to ≈ 5.3 Pa·s with 3.5 wt% oil added. At first glance, this finding is in contrast to Gu et al. who could only detect an increase in dynamic viscosity at oil concentrations above 10 % (Gu et al., 2009). However, in that study, soy protein isolate was used for gel formation. Once more, these differences might be explained by a different sensitivity of pea and soy protein gels to incorporate active (or inactive) fillers. In Fig. 5, the results of the amplitude tests of the different samples are shown. Furthermore, the addition of oil increases the ratio of protein to water, which leads to an increase in the maximum structuring speed of the developing protein network (Klost & Drusch, 2019).

The storage modulus and loss modulus increase with increasing oil content. The crossover point increased from ≈ 5 Pa for the yogurt alternative without oil (empty symbols) to ≈ 8 Pa with 1.5 wt% oil (half-filled symbols) and 14 Pa with 3.5 wt% oil (filled symbols). Corresponding to that, the LVE becomes longer with increasing oil content. In summary, thus, the yogurt alternatives show higher gel strength and can be characterized as more deformable and more withstanding to higher stresses when the oil content is higher (Adams et al., 2004). The PPI used to stabilize the oil droplets interacts with the PPI of the gel matrix. As a result, the oil droplets are incorporated into the gel network as an active filler, which strengthens the gel network (Dickinson & Chen, 1999; Wiedenmann et al., 2018).

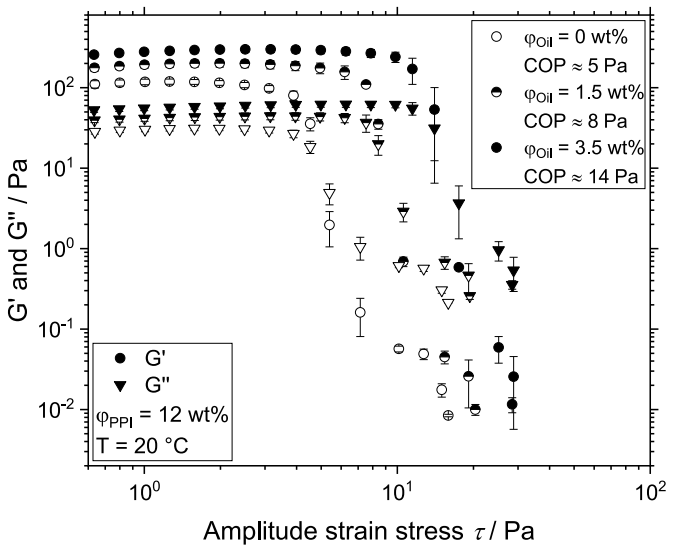

Fig. 5. Amplitude tests of yogurt alternatives produced without the addition of oil and with 1.5 wt% and 3.5 wt% canola oil, measured at 20 °C with a circular frequency of 1 rad·s^{-1}. The values of the crossover points (COP) can be found below the point labels in the legend to the right.

3.3. Influence of MGP and oil addition on the tribological properties of fermented pea protein gels

In order to evaluate whether the addition of MGP or oil to pea protein gels improves the lubrication properties of the yogurt alternatives, tribological analyses of the samples were conducted. Malone et al. (2003) found that fat perception of emulsions is related to lubrication in the speed range of 1–30 mm·s^{-1}. Consequently, the range shown up to 30 mm·s^{-1} is the range of interest for yogurt creaminess perception (Malone et al., 2003). Chojnicka-Paszun et al. (2012) have demonstrated in their research that the perception of creaminess is contingent upon the friction coefficient being below 100 mm·s^{-1} (Chojnicka-Paszun et al., 2012). It was expected that the addition of MGP would reduce the friction coefficient. The addition of particulate MGP allows rolling between the tongue and plate or rotor and stator and may therefore reduce the friction coefficient (Sarkar et al., 2017). The results, presented as

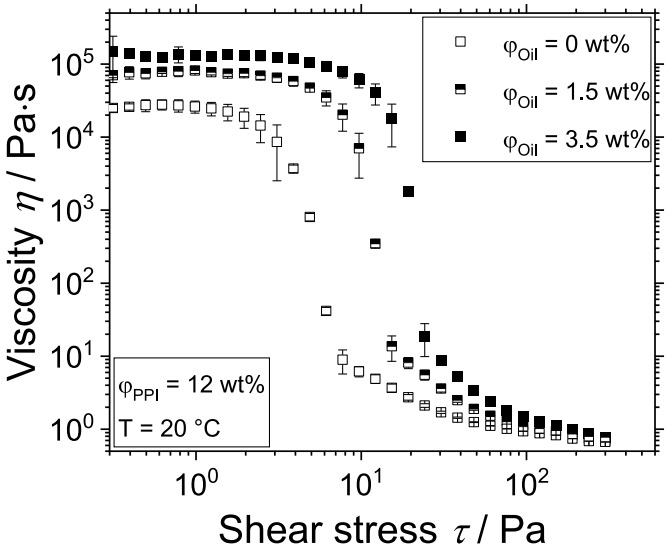

Fig. 4. Flow curves of yogurt alternatives produced without the addition of canola oil and with 1.5 wt% and 3.5 wt% oil, measured at 20 °C.

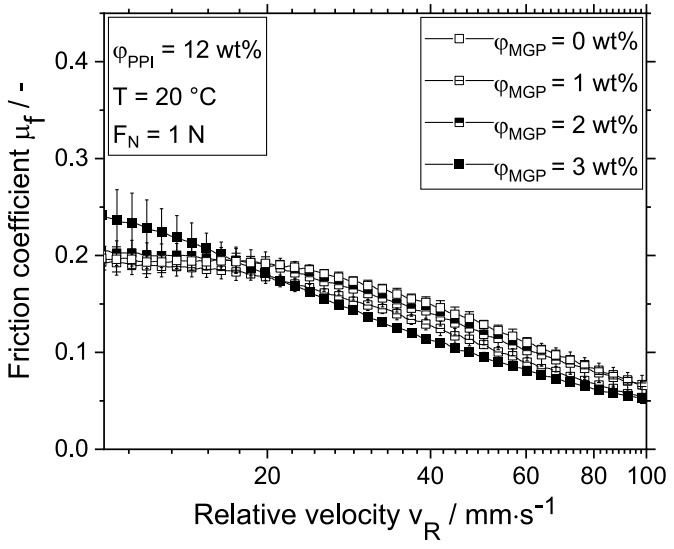

Fig. 6. Tribological measurements of yogurt alternatives produced without the addition of microgel particles and with 1 wt%, 2 wt% and 3 wt% microgel particles, measured at 20 °C, using PDMS pins and a glass ball as tribo-pairs.

Stribeck curves for the yogurt alternatives without and with 1 wt%, 2 wt % and 3 wt% MGP are shown in Fig. 6. The extended Stribeck curves are presented in Supplementary Material Fig. 1.

The relative velocity is plotted from 10 to 100 mm·s^{-1}, reflecting velocities relevant to swallowing (Grebe, 2017). Fig. 6 shows that all vegan yogurt alternatives displayed boundary (v_R = 10–20 mm·s^{-1}) and mixed regimes (v_R = 20–100 mm·s^{-1}) (Kew et al., 2021). The highest friction coefficient of ≈ 0.25 was measured at a relative velocity of 10 mm·s^{-1} for the sample containing 3 wt% MGP (filled squares). Above a relative velocity of 20 mm·s^{-1}, all curves are nearly parallel and decrease continuously with increasing velocity. Looking at a velocity of 40 mm·s^{-1}, differences between the samples seem to be observable. Taking into account the standard deviations, these are, however, statistically not significant. Therefore, no influence of MGP concentration on the lubrication properties in the absence of oil can be detected. It is known from literature that PPI forms aggregates that prevent mutual rolling and may even increase the friction coefficient (Zembyla et al., 2021). At a PPI concentration of 12 wt%, it can be assumed that such aggregates are indeed formed. Their size might probably be larger than the size of the MGP covering the lubricating effect of the MGP. It has been shown in the literature that the lubricating properties of dairy products as well as dairy substitutes can be modulated by the addition of various fat or oil contents (Laguna et al., 2017; Ningtyas et al., 2017; Saavedra Isusi et al., 2023). An increased oil content results in a lower coefficient of friction regardless of the tribological method selected (Nguyen et al., 2017; Sonne et al., 2014; Wijk & Prinz, 2005). The investigated pea protein gels were subjected to tribological measurements in order to confirm this finding. Fig. 7 shows the corresponding Stribeck curves of the yogurt alternatives without and with 1.5 wt% and 3.5 wt% oil. The extended Stribeck curves are presented in Supplementary Material Fig. 2.

The curves initially run in the boundary regime at a constant friction coefficient μ which is ≈ 0.20 for the yogurt alternative without added oil (empty squares), ≈ 0.16 with 1.5 wt% oil (half-filled squares) and ≈ 0.13 for 3.5 wt% oil (filled squares). At a relative velocity of approximately 20 mm·s^{-1}, the curves start to tilt, which corresponds to the onset of the mixed regime. In the relative velocity range relevant to creaminess perception (1–30 mm·s^{-1}) (Malone et al., 2003), the friction coefficient decreases with increasing oil content, as previously shown for dairy products. As a consequence, the yogurt alternative will appear creamier as previously investigated. The addition of oil leads to the formation of an interfacial film that prevents the tongue and palate from touching and allows sliding (Sonne et al., 2014). Increasing the oil concentration increases the amount of oil between the palate and tongue, allowing the system to be better lubricated (Baier et al., 2009). However, apart from the oil concentration, the oil droplet size can affect the friction and lubricating properties of emulgels (Wijk & Prinz, 2005). As shown in Fig. 1, the droplet size distributions of the two emulsions are very much alike. Therefore, it can be ruled out that the observed effect is due to the oil droplet size. This means that the decrease in the friction coefficient is only caused by the oil concentration.

3.4. Targeted modulation of yogurt alternatives based on pea protein by the addition of MGP and oil

A high protein content in yogurt is often the cause of product complaints such as increased firmness due to a firm gel structure and a less creamy mouthfeel (Janhøj et al., 2006; Jørgensen et al., 2019). Despite a lack of systematic research in this field, it can be assumed that also high protein vegan yogurt alternatives will be characterized by those sensory attributes. Thus, it is of interest to investigate how the corresponding texture properties can be modulated in vegan alternatives. In order to do so, the results obtained so far were used and further assumptions on texturing strategies in pea protein-based yogurt alternatives were made. It was shown above that oil can be used to improve the lubricating properties, i.e. to reduce the friction coefficient in fermented pea protein gels with high protein concentration. However, this can only be achieved at the expense of a stronger gel network due to the active filler effect of the oil droplets. In contrast, MGP can be used to weaken the pea protein gel network, and they do not affect the friction coefficient. Thus, it is assumed that the targeted addition of MGP to an oil-rich yogurt alternative will weaken the gel structure while maintaining the improved lubricating properties. In order to test this assumption, three yogurt alternatives were prepared with and without oil as well as with and without MGP. The flow curves are shown in Fig. 8.

As expected, the addition of 1.5 wt% oil shifts the yield point to higher shear stress of the yogurt alternative from ≈ 1.9 Pa (empty squares) to ≈ 3.8 Pa (half-filled squares), respectively. The addition of 3 wt% MGP results in a lower yield point of ≈ 3.0 Pa (filled squares) compared to 3.8 Pa in the sample with oil but without added MGP. This means that, indeed, MGP act as an inactive filler in the protein network even when oil is present. Thus, MGP can be used to weaken the gel

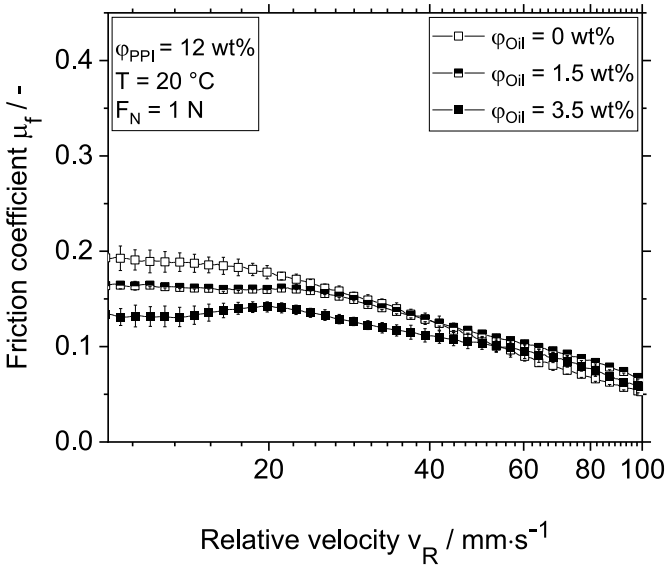

Fig. 7. Tribological measurements of yogurt alternatives produced without the addition of canola oil and with 1.5 wt% and 3.5 wt% canola oil, measured at 20 °C, using PDMS pins and a glass ball as tribo-pairs.

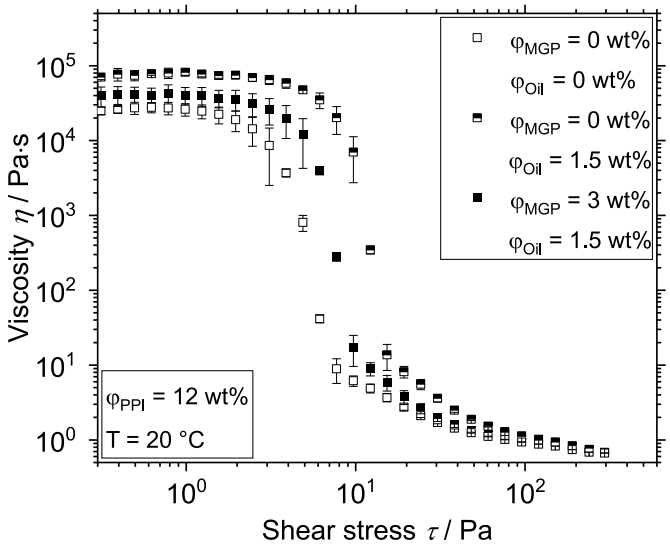

Fig. 8. Flow curves of yogurt alternatives produced with different MGP concentrations and without or with the addition of 1.5 wt% canola oil, measured at 20 °C.

structure of emulgels. Additionally, the Stribeck curves resulting from the tribological analysis of the three samples are shown in Fig. 9. The extended Stribeck curves are presented in Supplementary Material Fig. 3.

A constant friction coefficient between 0.15 and 0.2 up to a speed of ≈ 20 mm·s^{-1} (end of boundary regime) can be observed for all samples. The friction coefficient then decreases to 0.05 in the mixed regime (Kew et al., 2021). In the boundary regime, the friction coefficient of the oil-rich samples is lower than that of the yogurt alternative without oil. This is in line with the results shown in Fig. 7 and meets the expectations. When comparing the friction coefficient of the two samples containing oil and taking into account the standard deviations, it can be observed that the addition of MGP does indeed not affect the friction coefficient. Thus, the previously made assumption can be entirely confirmed: The addition of MGP to the pea protein-based yogurt alternative with 1.5 wt% oil can weaken the gel structure without affecting the friction coefficient, which is reduced by the addition of oil. Targeted modulation of structure and texture is possible.

3.5. Influence of oil content on the color of a yogurt alternative

Additionally, to the rheological and tribological characterization of the yogurt alternatives, the color of the samples was studied. Color is another sensorial aspect in which vegan yogurt alternatives often differ from their dairy benchmark. In contrast to cow's milk-based yogurt (L*, the pea protein-based yogurt alternative exhibits a more brownish or yellowish color (Milovanovic et al., 2020). Thus, the vegan alternative does not align with the expectations of consumers who mostly desire the optical properties known from dairy products.

It is well established that a product that fails to meet consumer expectations is associated with lower overall acceptance (Zellner et al., 2004). Therefore, it was investigated how far the strategies used to modulate the protein gel's texture can also be applied to influence the color, and thus possibly enhance the appeal, of the pea protein-based yogurt alternative. For this reason, the three yogurt alternatives investigated above were characterized by their color according to the CIE Lab system. The measured L*a*b* values are presented in Table 2.

The literature shows that as the oil concentration increases and the droplet size decreases, emulsions appear to become whiter and lighter in color (Chantrapornchai et al., 1998). The droplet size distributions of the emulsions were very much alike (compare Fig. 1 B), thus indicating

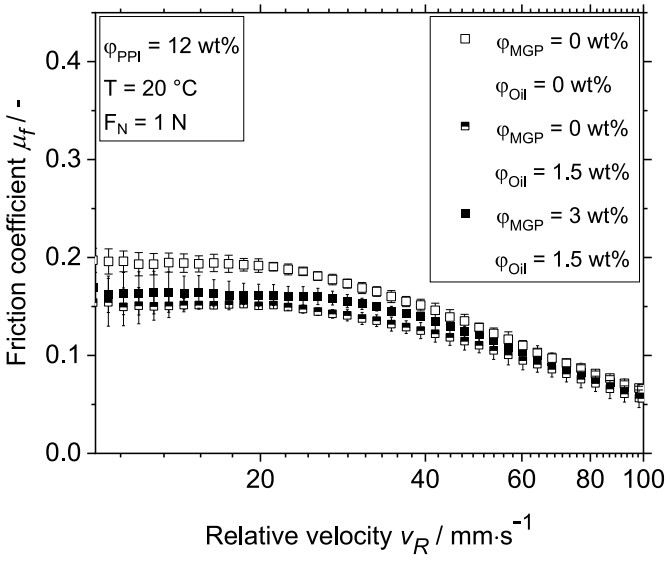

Fig. 9. Tribological measurements of yogurt alternatives produced with different MGP concentrations and without or with the addition of 1.5 wt% canola oil, measured at 20 °C, using PDMS pins and a glass ball as tribo-pairs.

Table 2
L*a*b*-values of yogurt alternatives without added canola oil and MGP, with added MGP or with added canola oil.

	0 wt% MGP / oil	3 wt% MGP	1.5 wt% oil	3.5 wt% oil
L*-value	74.6 ± 0.4	76.0 ± 0.6	80.1 ± 0.1	82.4 ± 0.1
a*-value	2.7 ± 0.0	2.2 ± 0.1	2.5 ± 0.1	2.7 ± 0.0
b*-value	17.1 ± 0.1	17.4 ± 0.2	16.1 ± 0.0	15.1 ± 0.1

that the effect of oil droplet size on coloration is negligible. Therefore, any changes in color perception should only be caused by the oil concentration.

A higher L*-value indicates a lighter or whiter color, as perceived by the human eye. It can be observed that the L*-value increases in conjunction with an increase in oil content, but there is no change in the L* value when MGP are added (Table 2). This indicates that the hypothesis can be corroborated, namely that the yogurt alternative appears optically whiter with increasing oil content. The a*-value denotes the color range from green with a value of −a* to red with a value of +a*. These variables exhibit only minimal variation and are therefore not considered further. The b*-value represents the colors blue (denoted by a value of −b*) and yellow (denoted by a value of +b*). The incorporation of 3 wt% microgel particles resulted in a marginal elevation in the b* value. However, no discernible alterations were evident when the yogurt alternative was assessed without the incorporation of MGP, taking into account the standard deviation. The addition of oil resulted in a reduction of the b*-value. This indicates that the yogurt alternative exhibits a diminished yellow hue with increasing oil content. In conclusion, it can be stated that the addition of oil results in a lighter or whiter appearance for the pea protein-based yogurt alternative, with a reduction in the yellow hue.

4. Conclusions

The objective of this study was to investigate the effects of MGP addition and canola oil addition on the texture and optical properties of pea protein-based yogurt alternatives. Rheological characterization indicated that MGP acted as inactive fillers, weakening the gel network. According to expectations, the addition of MGP decreased both the yield stress and the length of the linear viscoelastic region, demonstrating a reduction in gel strength and elasticity. In contrast, canola oil acted as an active filler, increasing the strength of the gel network and extending the LVE region. This contrasting behavior highlights the different mechanisms by which MGP and oil interact with the pea protein matrix. Tribological analysis showed that the addition of MGP did not significantly change the friction coefficient, likely due to the dominant effect of the PPI aggregates. However, the addition of oil consistently reduced the friction coefficient and improved lubrication. The optical properties of the pea protein-based yogurt alternatives were improved with oil addition, resulting in a brighter and less yellow appearance. In summary, targeted modulation of pea protein-based yogurt alternatives through the strategic addition of MGP and oil is possible. MGP can reduce gel firmness without compromising lubricity, while oil leads to firmer gels but also improves the lubricity. The addition of pectin-based MGP in combination with oil offers a promising modulation approach to design plant-based yogurt alternatives that better mimic the creamy mouthfeel and visual aspect of traditional yogurt, resulting in products that are more in line with consumer expectations.

CRediT authorship contribution statement

Désirée Martin: Writing – original draft, Visualization, Validation, Methodology, Investigation, Formal analysis, Conceptualization. **Eva Müller:** Validation, Investigation, Formal analysis. **Ulrike S. van der Schaaf:** Writing – review & editing, Supervision, Project administration, Funding acquisition, Conceptualization.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodhyd.2025.111595.

Data availability

Data will be made available on request.

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