J Food Sci Technol (July 2023) 60(7):2012–2022
https://doi.org/10.1007/s13197-023-05735-x

ORIGINAL ARTICLE

Effect of retrograded starch with different amylose content on the rheological properties of stored yogurt

Marco A. Dircio-Morales[1,2] · Gonzalo Velazquez[1] · Israel Sifuentes-Nieves[2] · Pamela C. Flores-Silva[2] · Heidi A. Fonseca-Florido[2] · Guadalupe Mendez-Montealvo[1]

Revised: 8 March 2023 / Accepted: 17 March 2023 / Published online: 26 March 2023

Abstract Resistant starch (RS) promotes health benefits; however, when added to foods, it could change the rheological properties. The effect of adding different concentrations (2.5, 5, 7.5, and 10%) of retrograded corn starch with 27% (RNS) or 70% (RHS) amylose content on the properties of yogurt was evaluated through measurements of flow behavior and gel structure. Syneresis and resistant starch content were also assessed. Results were analyzed using multiple regression to describe the effect of starch concentration and storage time on the properties of yogurt added with RNS or RHS. Syneresis was reduced, RNS reinforced the structure increasing the water absorption capacity and the consistency index; meanwhile, RHS provided a yogurt containing up to 10 g of RS in 100 g of sample, allowing obtaining a functional dairy product. Creep-recovery test showed that adding RNS or RHS favored the matrix conformation, and the yogurt samples were able to recover. The final product behaved like a solid material with a firmer and more stable gel structure, resulting in a strengthened gel without weakening the yogurt structure, showing a characteristic like Greek-style or stirred yogurt depending on the type and concentration of retrograded starch.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s13197-023-05735-x.

✉ Guadalupe Mendez-Montealvo
cmendez@ipn.mx

1 Instituto Politécnico Nacional, Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Cerro Blanco No. 141, Col. Colinas del Cimatario, C.P. 76090 Santiago de Querétaro, Querétaro, Mexico

2 Centro de Investigación en Química Aplicada, Blvd. Enrique Reyna No. 140, C.P. 25253 Saltillo, Coahuila, Mexico

Keywords Resistant starch · Yogurt · Viscosity · Creep-recovery · Amylose · Syneresis

Abbreviations

NS	Normal corn starch
HSVII	High amylose corn starch
RetS	Retrograded starch
RS	Resistant starch
RNS	Retrograded corn starch with 27% amylose
RHS	Retrograded corn starch with 70% amylose
RS3	Resistant starch type 3
CY	Control yogurt
G′	Storage modulus
G″	Loss modulus
LVR	Linear viscoelastic region
k	Consistency index
n	Flow behavior index
Tan δ	Loss tangent
η*	Complex viscosity

Introduction

Rheology studies the flow and deformation of food systems to predict the quality, texture, processing, and storage conditions (Zhong 2019). In this sense, texture is one of the most important properties of yogurt quality as it is related to the sensory perception. Health benefits attributed to the consumption of yogurt are mainly related to the presence of probiotics (Allgeyer et al. 2010). However, still there is a need for ingredients that can be included in the yogurt formulation, such as prebiotics, to make it a healthier functional food product.

Resistant starch (RS) is a fraction of starch that resists enzyme digestion, and it is not absorbed in the small intestine (Langkilde et al. 2002; Ashwar et al. 2016) but it is fermented in the colon promoting the growth of beneficial colonic microbiota (Wang et al. 2015). RS is also a functional ingredient that improves the texture in the final product due to its water holding capacity (Baixauli et al. 2008; Fuentes-Zaragoza et al. 2010). Among RS types, resistant starch type 3 (RS3), obtained from retrograded starch (RetS), has been recognized for its fast retrogradation of amylose molecules which promote a rapid decrease of the digestibility mainly because of the rearrangement of the amylose and amylopectin chains into a different and more ordered structure that takes place after the gelatinization and cooling process (Hoover et al. 2010; Wang et al. 2015).

The rheological properties of yogurt added with RS have been previously reported. Rezaei et al. (2017) investigated the effect of resistant starch and β-glucan on the steady and dynamic rheological properties of frozen soy yogurt blends. The authors found that adding RS enhanced the pseudoplasticity and increased the storage modulus (G') and loss modulus (G″), resulting in yogurt with liquid viscoelastic behavior. Meanwhile, He et al. (2019) reported that 2 types of resistant starches affected the progress of gelation, increased the viscosity, and maintained high viable counts of *Lactobacillus*, which improved the quality and the probiotic effect of yogurt. We hypothesized that RS3 obtained from normal or high amylose starches determines the rheological properties of the yogurt and it may play an important role in the final characteristics of the yogurt as an additive or as a functional ingredient. On this regard, previous studies have investigated the use of native or modified starches from different sources to modify the texture parameters of different types of yogurt; however, most of them tested concentrations up to 2% (Gustaw et al. 2011; Rezaei et al. 2018; Flores-Mancha et al. 2021; Szołtysik et al. 2021; Jia et al. 2022; Li et al. 2023). In our work, we used a starch concentration five times higher aiming to obtain a functional product with different consistency from stirred to Greek-style yogurts, reaching different preferences of the consumer. Our main interest was to study the effect of adding retrograded starch from normal (RNS) and high amylose (RHS) starches at different concentrations on yogurt examining the morphological and rheological properties after storage for 0, 7, and 14 days.

In this study, the changes in the rheological properties of yogurt were assessed after adding retrograded corn starch at concentrations up to five times higher than those reported in other studies. As described by Rezaei et al. (2018), fiber contributes to the modification of rheological and sensory characteristics of food, so, explaining how the rheological properties of yogurt are affected after adding retrograded starch could be relevant for dairy industry. To understand that effect, the content of resistant starch in the final product was assessed and a deep study of the rheological properties was carried out applying rotational and dynamic tests to characterize the flow characteristic of the system and to describe the viscoelastic properties of the system.

Materials and methods

Two types of corn starch were used. Normal starch (NS) and high amylose starch (HS-Hylon VII) were purchased from National Starch and Chemical S.A. de C.V. (Toluca, Mexico). The datasheet provided by the manufacturer indicates that the amylose content is 27 and 70% in the normal and the high amylose starches, respectively. To evaluate the effect of retrograded starch addition, Yoplait® brand natural commercial liquid yogurt was used.

Retrogradation of starch

The methodology reported by Soler et al. (2020) with slight modifications was followed to obtain retrograded starch (RetS). The starch suspension was heated at 120 °C (15 psi) during 30 min and then dried at 80 °C for 48 h using an air forced-convection oven. The powdered retrograded starch from normal (RNS) and high amylose (RHS) were stored until used.

Scanning electron microscopy

The dry sample, either from retrograded starch or yogurt, was placed on the aluminum holders using a conductive double-sided carbon tape to examine the morphological features using a scanning electron microscope (Phenom Pro, Phenom-World., Netherlands) at an accelerating voltage of 5 kV. Images were taken at 1500 X magnification.

Resistant starch

A resistant starch assay kit from Megazyme (Ireland) was used to determine resistant starch in yogurt (added with and without retrograded starch). Yogurt samples were dried overnight at 50 °C in an oven (Binder model BD 56), ground, and passed through a 250 µm mesh. The manufacturer's instructions were followed to measure the resistant starch content.

Syneresis

To evaluate the syneresis, the method reported by Keogh and O'Kennedy (1998) was followed.

Rheology measurements

Before measuring the rheological properties, a standardized procedure was established to prepare the samples. First, the starch was added to the yogurt and manually stirred until obtaining a homogeneous mixture. The blend was cooled for 24 h at 5 °C. After storage, the mixture was poured into Petri dishes, stirred 10 times, and settled for 15 min in a refrigerator at 5 °C. Finally, a sample was poured on the rheometer plate to repose for 5 min. This reposing time prevented the rheology memory of the sample. The rheological properties of the samples were measured in yogurt samples added with 2.5, 5, 7.5, and 10% of retrograded starch (RNS or RHS) using an Anton Paar rheometer (Physica MCR 101) with a plate-plate geometry of 50 mm and a gap of 1 mm. The measurement was carried out in samples stored for 0, 7, and 14 days at 4 °C.

Flow curve

The flow behavior of yogurt added with retrograded starch was measured using the flow curve test. Two measurement cycles, upward and downward from 0.03 to 300 s^{-1}, were applied for the test. The equation of the power-law model of Ostwald-de Weale was used to describe the flow curves. The consistency index (k, Pa s^n) and flow behavior index (n, dimensionless) were calculated from the flow curve test performed in triplicate.

Viscoelastic properties

The viscoelastic properties were measured by oscillatory tests using the same rheometer and geometry as described above. The parallel plates were covered with mineral oil to avoid water evaporation during the test. The linear viscoelastic region (LVR) was determined using deformation sweeps at 10 °C, and all tests were run at a frequency value of 1 Hz. Once the LVR was found, the rheometer was programmed for running a frequency sweep (0.05 to 5 Hz) with a constant strain value of 0.07%. The storage modulus (G′, Pa), loss modulus (G″, Pa), and loss tangent (Tan δ) were evaluated for each test. The test was performed in triplicate.

Creep-recovery test

Creep-recovery tests were carried out following the same standardized procedure to prepare the samples. For this test, a constant stress of 1 MPa was applied for 5 min and the recovery was measured for 10 min.

Data analysis

Data were analyzed by multivariable polynomial regression to describe the effect of storage time and starch concentration on the properties of yogurt with each type of starch. In the case of resistant starch content, the effect of amylose content and starch concentration was assessed up to 14 days of storage. Data were fitted using a second-order polynomial equation in Design Expert® v10 (Stat-Ease, Inc., Minneapolis, MN, USA).

Results and discussion

Health benefits associated to dietary fiber consumption have promoted its incorporation into foods and the resistant starch has been considered as a fiber. The fraction of RS in retrograded starches with different amylose content was 8.01 ± 0.26 and 8.16 ± 1.05% for RNS and RHS, respectively. These starches were added at 2.5, 5, 7.5, or 10% (w/w) to yogurt. The nutritional label of the commercial yogurt indicates that it contains 11.1 g of available carbohydrates, from which, 8.4 g were added; however, it does not report dietary fiber content. Fuentes-Zaragoza et al. (2010), Lamsal & Faubion (2009) and Jia et al. (2022) mentioned that some fibers have functional properties such as water absorption capacity, swelling, and gel formation. These properties allow improving the overall quality, favoring the sensory characteristics, and enhancing the stability of emulsions when added to several types of food including dairy products (Al-Sheraji et al. 2013; Li et al. 2023). This study focused on assessing the effect of the type and concentration of retrograded starch on the properties of commercial yogurt.

Microstructure of retrograded starches and Yogurt samples

The material obtained after starch retrogradation was a fine white power that was easily incorporated into the yogurt. Supplementary Figure S1A shows the micrographs of retrograded starches while the samples of yogurt added with RNS or RHS are shown in Supplementary Figure S1B. There were important differences in the structure of control yogurt (CY) compared to the yogurt added with RNS or RHS at 7.5%. CY showed a continuous network while the yogurt added with RHS showed covered caseins micelles probably acting as a reinforcing material. The entanglement in the structure seems more continuous than that observed after adding RNS. RHS promoted a denser network meanwhile RNS strengthened the matrix as it could be acting as a water structuring agent (Crispín-Isidro et al. 2015).

Regression analysis

Table 1 shows the calculated coefficients for each term in the second-order polynomial equation for RS as a function of concentration and amylose content while Table 2 shows the calculated coefficients for n, k, syneresis, G′, and Tan δ, as well as the R^2 value, adjusted R^2, and adequate precision for yogurt added with RNS and RHS as a function of concentration and storage time. The bigger the absolute value of the regression coefficient in the model, the higher the effect on the measured parameter. R^2 indicates the percentage of variation explained by the factors affecting the measured parameter. In this study, some of the evaluated parameters did not have a high R^2 value for the fitting of the regression model. Adjusted R^2 considers the impact of additional independent variables on the predicted values of the model. A difference between the adjusted R^2 and the R^2 less than 0.2 is usually recommended. In general, there was a reasonable agreement between the R^2 and adjusted R^2 values as shown in Table 2. The adequate precision allows evaluating the significance of a model. Adequate precision values higher than 4 are desirable. In this analysis, the lowest value for adequate precision was 3.874. In general, the values for R^2, adjusted R^2, and adequate precision suggest that the second-order polynomial equation can be used to describe the effect of the studied factors on the measured properties of commercial yogurt. Table 2 shows the statistical parameter (*p*-value) obtained from the ANOVA of the fitting of the regression model. *P*-values lower than 0.05 indicate a significant effect.

Table 1 Final equations and statistical parameter (*p*-value) for resistant starch (RS) in yogurt added with RNS or RHS at different concentrations

Factor	Coefficient/parameter	*p*-value
Intercept/model	5.931	< 0.001
A-starch concentration	2.655	< 0.001
B-amylose content	2.463	< 0.001
AB	1.964	< 0.001
A^2	− 2.817	< 0.001
R^2	0.968	
Adjusted R^2	0.963	
Adequate precision	35.317	

Resistant starch content

It is well known that fiber consumption is lower than that recommended by the World Health Organization (WHO), that is why it is important to include dietary fiber as an ingredient into food formulations. The popularity of dairy products added with prebiotics has increased as consumers are more aware of their health benefits (Allgeyer et al. 2010;

Table 2 Final equations and statistical parameter (*p*-value) for index flow behavior (n), index consistency (k-Pa s^n), syneresis (%), storage modulus (G′), and loss tangent (Tan δ) for stored yogurt added with RNS or RHS at different concentrations

Term/parameter	Syneresis (%)		k (Pa s^n)		G′ (Pa)		Tan δ	
	Coefficient	*p*-value	*Coefficient*	*p*-value	*Coefficient*	*p*-value	*Coefficient*	*p*-value
RNS								
Intercept/Model	46.07	< 0.001	7.9	0.001	319.41	0.226	0.1691	0.007
A-Starch concentration	− 12.64	< 0.001	4.31	< 0.001	233.66	0.020	− 0.0075	0.001
B-Storage	− 2.63	< 0.001	0.9073	0.106	25.96	0.727	− 0.001	0.504
AB	− 0.8807	0.2539	1.07	0.204	6.14	0.953	0.0035	0.109
A^2	6.72	< 0.001	− 0.4427	0.659	− 14.83	0.918	0.0061	0.050
B^2	2.23	0.0215	− 1.62	0.077	− 93.99	0.470	− 0.0038	0.148
R^2	0.925		0.892		0.488		0.787	
Adjusted R^2	0.9162		0.825		0.204		0.669	
Adequate precision	28.388		11.369		4.032		7.937	
RHS								
Intercept/Model	45.91	< 0.001	8.87	0.007	186.17	< 0.001	0.1746	0.234
A-Starch concentration	− 11.45	< 0.001	3.25	< 0.001	131.82	< 0.001	− 0.0015	0.425
B-Storage	− 2.74	< 0.001	0.2228	0.604	36.41	0.018	− 0.0033	0.066
AB	− 2.81	0.0014	− 0.0663	0.912	48.68	0.023	− 0.0006	0.806
A^2	8.94	< 0.001	− 1.4	0.119	39.55	0.144	0.0048	0.161
B^2	− 0.0717	0.9434	− 2.88	0.003	− 62.81	0.018	− 0.0028	0.346
R^2	0.909		0.878		0.922		0.483	
Adjusted R^2	0.879		0.811		0.879		0.195	
Adequate precision	24.617		11.634		14.240		3.874	

Mwizerwa et al. 2017). Resistant starch (RS) is not digested and when reaching the colon, it is fermented as the dietary fiber does and this has been associated with weight management and health benefits (Bello-Perez et al. 2020). Figure 1 shows the content of RS as a function of amylose content and starch concentration. The RS content was measured in yogurt samples stored for 14 days at 4 °C.

Table 1 shows that all terms in the second-order polynomial model had a significant effect on the RS content ($p < 0.05$). An R^2 value of 0.971 and adjusted R^2 of 0.948 implies a good fitting and the value of adequate precision indicate that the polynomial model can be used to describe the effect of amylose content and starch concentration on the content of resistant starch in yogurt added with retrograded starch.

The amount of RS in yogurt increased after adding retrograded starch. This increase was more noticeable when RHS was added. It has been reported that retrograded amylose resists the action of digestive enzymes mainly because its well-organized parallel or elongated arrangement in the structure contributes to the RS fraction (Bello-Perez et al. 2020). Because of the low amylose content, RNS did not significantly increase the RS content in yogurt despite the added concentration. Mwizerwa et al. (2017) added 1% of modified cassava starch and reported values of RS similar to those found in this study. These results could have contributed to the high capacity of the matrix to retain water, favoring a better texture, and increasing the consistency of the yogurt, as it will be discussed further.

The RS values reached at 14 days of storage in the yogurt added with RHS make it feasible to be considered as a functional food with nutraceutical effects, providing from 2 to 10 g of RS for 100 g of sample. It has been reported that consuming RS results in several metabolic benefits including a healthy gut microbiota (Bello-Perez et al. 2020).

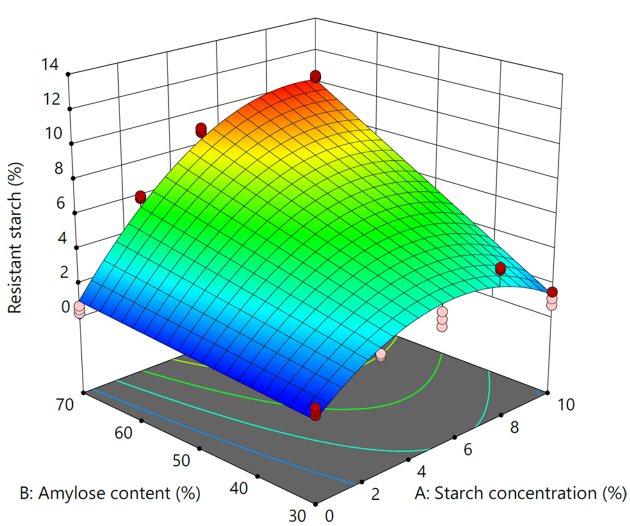

Fig. 1 Resistant starch content in yogurt added with RNS or RHS at different concentrations evaluated after 14 days of storage at 4 °C

Syneresis

Syneresis is a major visible defect (Sah et al. 2016) as the accumulation of liquid on the surface of yogurt gels affects the acceptability of the product. The effect of starch concentration and storage time on the percentage of syneresis for RNS and RHS is shown in Fig. 2a, b, respectively. The fitting of the second-order polynomial equation to the experimental data (Table 2) for the addition of both types of starch indicates that this property is affected mainly by the linear and the quadratic term of the starch concentration. Both terms had the highest absolute values and the lowest *p*-values (Table 2). The linear term was also significant for RNS and RHS. The R^2 value was at least 0.930 and the adequate precision shows that the model can be used to describe the effect of starch concentration and storage time on the syneresis of yogurt.

As shown in Fig. 2a, b, the percentage of syneresis decreased in all yogurt samples as the concentration of retrograded starch increased. The values at 14-days storage were slightly higher for the samples added with RNS. Interestingly, at each concentration, the syneresis percentage remained stable during storage for samples added either with NSR or HSR. After 14 days, the syneresis values were around 40%, lower than those reported by Codină et al. (2016) who added up to 2% of quinoa flour to yogurt. The authors mentioned that the increase of syneresis was attributed to an unstable protein network. In another study, Sah et al. (2016) reported syneresis values around 2% when pineapple peel powder and inulin were added to yogurt. In this study, the capacity of retrograded starch to adsorb and bind water improved the stability of the yogurt samples, favoring the continuity of the network and the gel conformation. This effect was more noticeable when RNS was added as lower values of RS were reached and a more structured matrix was observed in SEM results. A remarkable behavior was also observed when RHS was added. These samples showed a more compact structure as observed in SEM, higher RS content, and adequate water holding capacity. The syneresis values were superior to those reported by Crispín-Isidro et al. (2015) who found values from 2.6 to 9.2 g/100 g in reduced-fat yogurt added with agave fructans and inulin. The differences could be explained by the structural properties of the polysaccharides. The polymeric chains of inulin and agave fructans are smaller than those of starch leading to a less strengthened matrix or gel structure which results in higher syneresis values in our study. Another important factor is the capability of retrograded starch to interact with water. As reported by Jiang et al. (2019) and Wang et al. (2021), a high number of hydroxyl groups in the polymeric

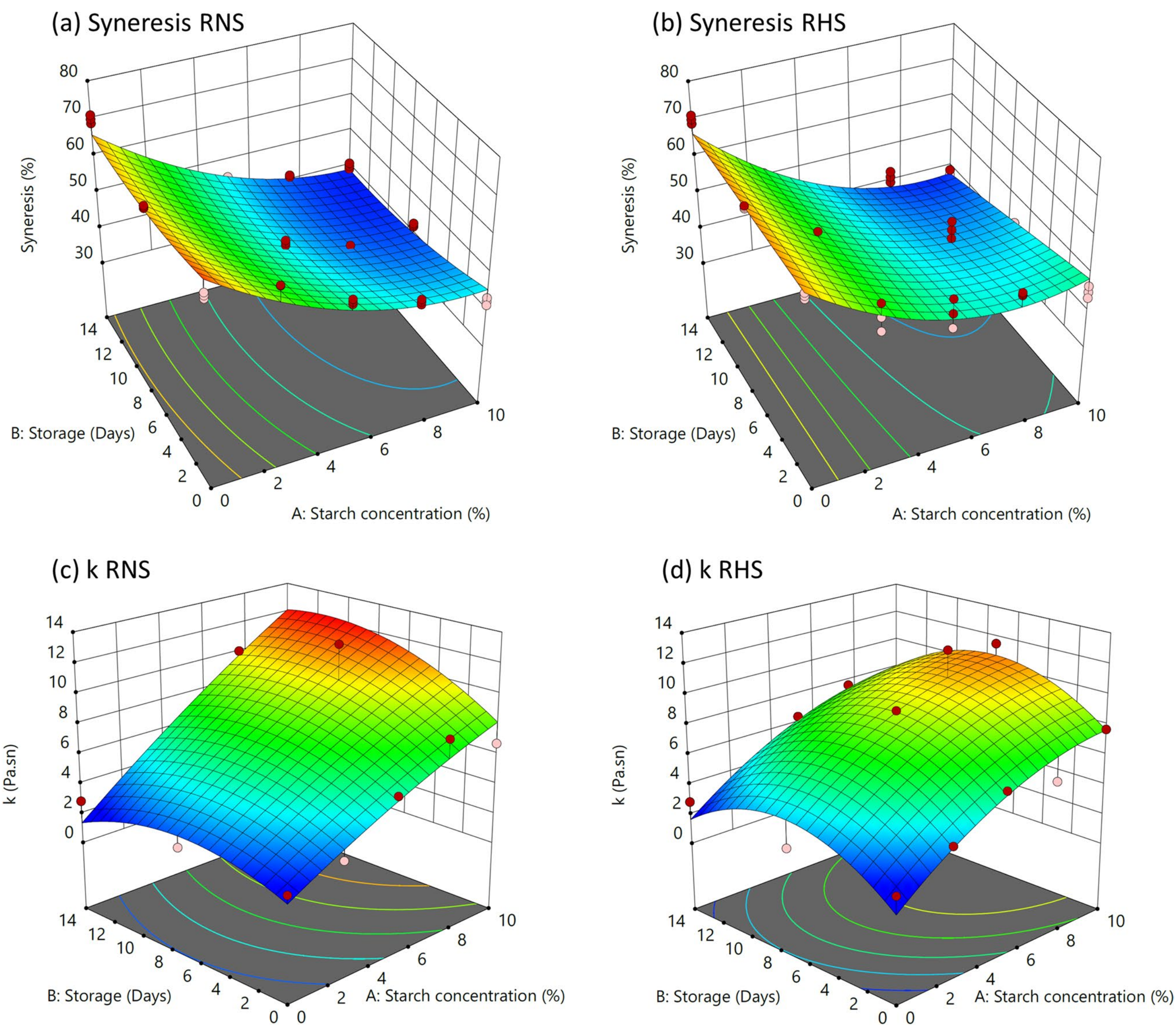

Fig. 2 Syneresis percentages and index consistency of yogurt samples added with RNS (**a**, **c**) or RHS (**b**, **d**), at different concentration evaluated for 14 days stored at 4 °C

chain of polysaccharides promotes the interaction with water molecules by hydrogen bonds, increasing the water holding capacity and reducing the syneresis. In this study, these interactions with water molecules allowed reducing about 60% the syneresis of liquid yogurt when retrograded starch was added. As most of the related studies are carried out with set yogurts, controlling the syneresis of liquid yogurts could be an important application of retrograded starches.

Rheological characterization

Knowing the rheological properties of food systems is important for quality control, processing, and acceptance by consumers, mainly because the rheological behavior of complex systems is affected by several factors including the concentration and interaction among the ingredients (Rezaei et al. 2017, 2018; Jia et al. 2022; Li et al. 2023). The interactions taking place between the yogurt components and the retrograded starch are the main interest of this study.

Flow curves

The flow behavior index (n) of yogurt added with retrograded starch with different percentages of amylose for all samples was lower than 1 (data no-shown), meaning a non-Newtonian fluid and describing a shear-thinning behavior. The viscosity decreased as the shear rate increased, although the dependence on the shear rate was more noticeable when RNS was added. In the case of RHS, the samples were more stable, and this behavior could be explained by the readily orientation of the lineal molecules (amylose) when the rotation tests are applied.

The consistency index *k,* as a function of storage time and starch concentration for yogurt added with RNS and RHS, is shown in Fig. 2c, d, respectively. The statistical analysis shown in Table 2 indicated that the *k* parameter was significantly affected by the linear term of the starch concentration when RNS was added to the yogurt. In the case of RHS, only the linear term of starch concentration and the quadratic term of the storage time had a significant effect on *k* ($p < 0.05$). The calculated R^2 value was 0.892 and 0.878 meanwhile the adequate precision values were 11.369 and 11.634 when RNS and RHS were added, respectively, showing that the polynomial model can be used to describe the effect of starch concentration and storage time on the *k* parameter. For RNS, as the added concentration and storage time increased, higher *k* values were found. The differences observed after adding RHS were less noticeable. Although there was a slight increase after 7 days of storage, the *k* values in samples stored for 14 days were similar to those observed at day 0 for all samples.

Compared to the control sample, an increase of around 4-folds and 3-folds was observed when 10% of RNS or RHS was added, respectively. As the initial sample was highly fluid, different textural characteristics like those of Greek-style or stirred yogurt, can be obtained by adding different concentrations of retrograded starch. The textural properties could also change depending on the type of retrograded starch (RNS or RHS). High values of *k* indicate better firmness of the yogurts because of the restriction in the molecular motion due to entanglements of polymeric chains in the gel matrix (Codină et al. 2016). The higher increase in the *k* values in yogurt added with RNS is due to the amylopectin content, as it could be more difficult for the polymeric chains to rearrange in the same direction as amylose does. Also, likely the linear chains of amylose hindered the development of a three-dimensional structure in the gel, resulting in better consistency (Rezaei et al. 2017, 2018). The results suggest that adding retrograded starch promotes a higher shear thinning behavior, resulting in properties that could be useful to stabilize particle suspensions processed at low shear rate conditions (Rayment et al. 1998; Dogan et al. 2013). As the concentration of retrograded starch increased, the consistency index increased due to the strengthening of the yogurt gel structure. Opposite results were reported by Witczak et al. (2020) when substituted inulin in potato starch gels.

Dynamic rheology: gel conformation

Gel formation is the basic phenomenon taking place during yogurt manufacturing (Sah et al. 2016). The yogurt gel firmness mainly depends on the strength of the three-dimensional network of milk proteins and, in this study, the retrograded starch. The amylose confers high stability reflected in the increase of storage modulus (G′) when RNS or RHS was added to yogurt (Fig. 3a, b). The ANOVA results (Table 2) for the fitting using the second-order polynomial equation shows that only the linear term of starch concentration had a significant effect on G´ when RNS was added; meanwhile, when RHS was added, the significant effects were the linear term of starch concentration and both the linear and the quadratic terms of storage as well as the interaction of both independent variables. Table 2 shows a R^2 value of 0.488 and 0.922 as well as adequate precision values of 4.032 and 14.24 for the fitting of G′ when RNS and RHS were added, respectively. G′ behavior suggests that the network formed by adding RNS at 10% favored the stability of yogurt as indicated by the low dependence on the frequency, exhibiting properties of a true gel as G′ was predominant over G″ (data not shown). The gel matrix was denser with longer stability, providing uniformity in the protein-carbohydrate matrix, and favoring the water retention capacity. At higher concentration, a better entanglement was reached in the yogurt as shown by the SEM micrographs (Supplementary Figure S1B), the syneresis values, and the behavior of G′. In this study, the G′ values increased from 59.06 Pa at 0 days in CY to a maximum of 914.25 Pa for samples added with 7.5% RNS at 7 days (Fig. 3a), and to 397.50 Pa at 14 days when RHS was added (Fig. 3b). These changes in storage modulus are attributed to different matrix properties conferred by the retrograded starches. RNS is a strong material that promotes water retention capacity as shown by the syneresis values; meanwhile, RHS did not increase the consistency of material noticeably, prevailing the entanglement and favoring the increase of the resistant starch amount, as previously mentioned.

Tan δ (Fig. 3c, d) is the ratio between loss and storage moduli. This parameter is used to describe the matrix characteristic as low values of Tan δ indicate more entanglement of the macromolecules participating in the structural organization of the system (Ferry 1980). Differences in Tan δ were observed between CY and the yogurt added with retrograded starch. In samples with RNS, the analysis of variance (Table 2) shows that only the linear term of starch concentration had a significant effect on the Tan δ values ($p < 0.05$). When RHS was added, the most influencing parameter was the linear term of storage time although the effect was not significant ($p > 0.05$). Table 2 shows R^2 values of 0.787 and 0.483 for RNS and RHS respectively. In both cases, the adjusted R^2 values were

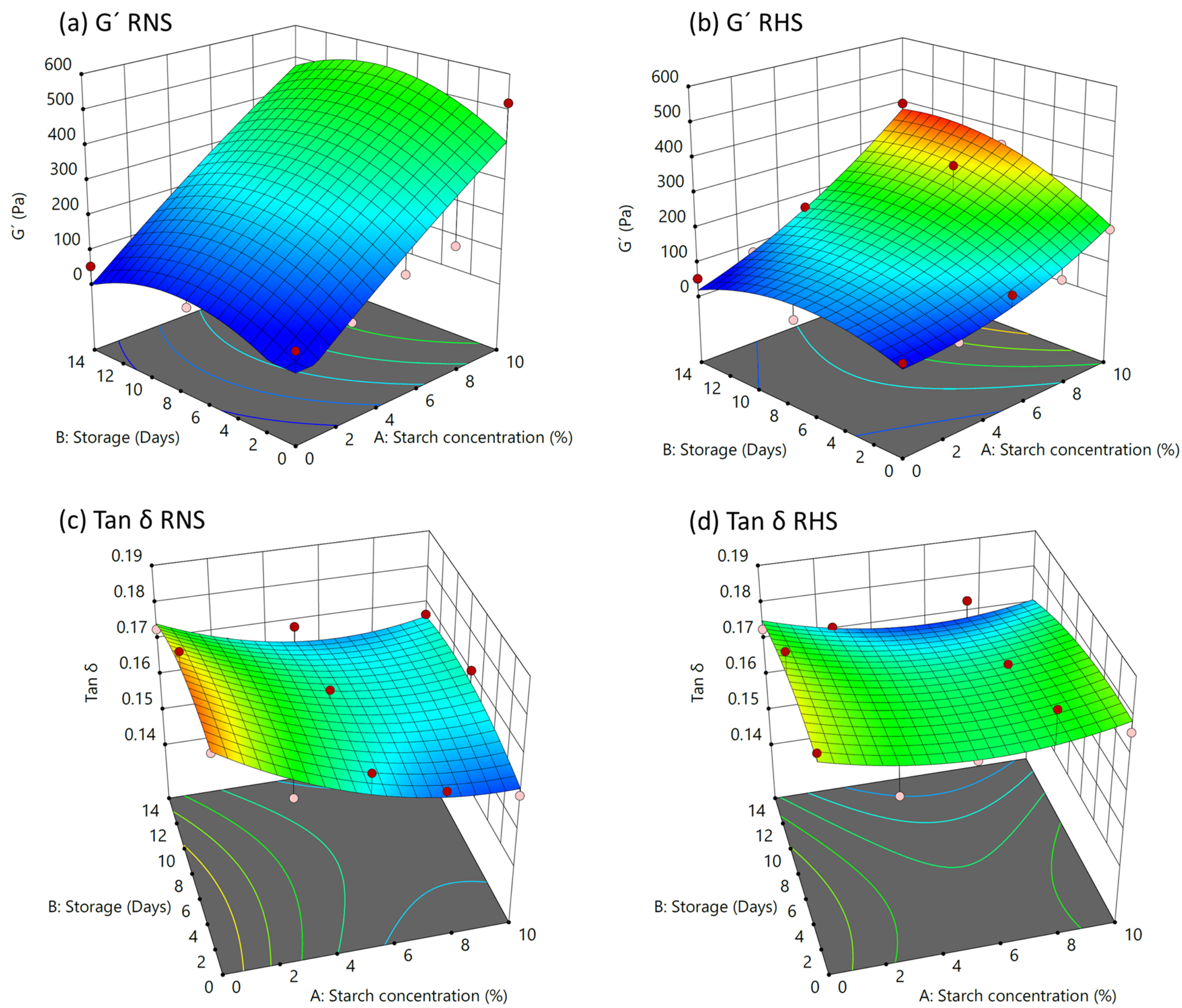

Fig. 3 Storage modulus (G′) and loss tangent (Tan δ) for yogurt samples added with RNS (**a**, **c**) or RHS (**b**, **d**) at different concentration evaluated for 14 days stored at 4 °C

in reasonable agreement with R^2. The sample added with 10% of RNS had the lowest values indicating that a higher amount of water retained favored the mobility in the matrix improving the entanglement. The stability also observed when RHS was added is explained by the low amylopectin content and the lower syneresis values reached indicating that the structure was more compact as shown in SEM images. As reported by Sah et al. (2016), loss tangent (Tan δ) decreased with storage time, indicating an enhanced network elasticity and a better rearrangement of the gel structure shifting toward a more solid-like behavior. The decreasing of Tan δ values reflect the elastic property of the yogurt samples, indicating that the solid-like character increased with the addition of retrograded starch (Dogan et al. 2013; Witczak et al. 2020; Jia et al. 2022), enhancing the texture of yogurt. This behavior suggests that retrograded starches promoted cross-linking between biopolymers in the yogurt (Wang et al. 2021), providing a yogurt with different texture as a function of the type of retrograded starch that is added.

The complex viscosity η* points to almost no change during storage maintaining the values around 19.11 Pa s. Samples added with RNS or RHS reached the highest values of 295.0 Pa s. at 7.5% after 7 days or 128.35 Pa s at 10% after 14 days, respectively. Compared to CY, there was an important increment of 15- and 6-folds for RNS and RHS, respectively, showing that the structural differences in yogurt promoted by the retrograded starches affected this behavior. The amount of amylopectin favored the water-binding property in the control samples increasing the complex viscosity. Witczak et al. (2020) and Li et al. (2023)

mentioned that the increase of the moduli values is related to the thickening properties of polysaccharides. This behavior was favored when the total solids increased, promoting the interaction among different components in the yogurt added with retrograded starch.

Creep-recovery test

Creep test is used in viscoelastic foods to obtain valuable information on the rheological parameters (Rao 2014) and it has been used to characterize gels (Tejada-Ortigoza et al. 2020). In this test, an instantaneous stress is applied, and the strain is observed over time to evaluate the properties of the material such as the capability to recover the original shape (Noosuk et al. 2005), this behavior can be described using the Maxwell model, allowing evaluating the creep-recovery parameters. The creep-recovery test helps to describe the internal structure of a system and the structural changes depending on the composition (Dogan et al. 2013; Jia et al. 2022). In this study, the creep-recovery test was applied to evaluate the texture and the stability of yogurt after adding RNS or RHS.

The gel structure in the yogurt is a result of casein aggregation by pH reduction and disulfide bonding between κ-casein and denatured whey proteins (Sah et al. 2016). In this study, the addition of retrograded starch contributed to the formation of the gel structure. The behavior of the creep-recovery response of yogurt added with RNS or RHS at different concentrations at 0-days storage is shown in Supplementary Figure S2. The highest deformation was observed for CY. As the concentration of retrograded starch increased, the deformation of the gel structure was lower suggesting that the matrix was more compact reaching a better entanglement of the biopolymers as demonstrated by the Tan δ values. This effect was more noticeable for RNS as less deformation and lower recovery were observed at higher concentrations, probably due to a better interaction between amylopectin or amylose and the yogurt constituents. As the concentration of retrograded starch increased, the yogurt became less spongy due to the presence of pores (Del Nobile et al. 2007). In all cases, the yogurt samples added with retrograded starch were able to recover, behaving like a solid material. Also, the gel structure was firmer and more stable as a function of the added concentration (Dogan et al. 2013; Jia et al. 2022).

The creep recovery curve of yogurt added with retrograded starch exhibited a non-linear deformation in the creep range. A similar trend was observed by Wang et al. (2021) who worked with dough added with insoluble fiber. In this study, the position and shape of the curves were strongly influenced by the addition of retrograded starch. The creep stage decreased when the concentration of RNS or RHS increased and, after removing the constant stress, the yogurt matrix partially recovered over time.

Conclusion

The effect of RNS or RHS on the gel structure of yogurt was assessed through rheological measurements. The yogurt matrix became more compact as the concentration of retrograded starch increased; however, different concentrations resulted in distinctive characteristics allowing tailoring the final properties of yogurt as shown by the consistency index. RNS reinforced the structure increasing the water absorption capacity and providing an important amount of resistant starch. Higher content of resistant starch was observed in yogurt with RHS. The yogurt added with retrograded starch was stable during storage as demonstrated by G′ and Tan δ results. The creep-recovery test showed that adding retrograded starches favored the matrix conformation in the yogurt increasing the resistance to deformation under stress conditions. This effect could lead to the strengthening of the gel without weakening the yogurt structure, providing a broad range of applications, modifying the texture of dairy food to meet the preferences of the consumers.

Retrograded starch improved the physicochemical properties of yogurt, favoring the syneresis and improving the RS content. The values of RS reached by RHS allowed obtaining a functional dairy product.

Acknowledgements We appreciate the financial support from SIP-IPN, COFAA-IPN and EDI-IPN.

Authors' contributions MADM: Methodology, investigation. GV: Visualization, formal analysis, review and editing of manuscript and fund acquisition. ISN: Review, research. PCFS: writing-review of manuscript. HAFF: Methodology, resources, review of manuscript. GMM: Conceptualization, writing original draft, review and editing of manuscript and fund acquisition.

Funding Secretaria de Investigacion y Posgrado del Instituto Politecnico Nacional; Consejo Nacional de Ciencia y Tecnología (CONACYT).

Data availability The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Code availability Not applicable.

Declarations

Conflicts of interest There is no conflict of interest to be declared by the authors.

Ethics approval The experimental procedures were conducted at CICATA-Qro-IPN under the ethical guides of this educational institution.

Consent to participate The authors consented to participate in this research project.

Consent for publication Not applicable.

References

Allgeyer LC, Miller MJ, Lee S-Y (2010) Sensory and microbiological quality of yogurt drinks with prebiotics and probiotics. J Dairy Sci 93:4471–4479. https://doi.org/10.3168/jds.2009-2582

Al-Sheraji SH, Ismail A, Manap MY et al (2013) Prebiotics as functional foods: a review. J Funct Foods 5:1542–1553. https://doi.org/10.1016/j.jff.2013.08.009

Ashwar BA, Gani A, Shah A et al (2016) Preparation, health benefits and applications of resistant starch—a review. Starch Stärke 68:287–301. https://doi.org/10.1002/star.201500064

Baixauli R, Salvador A, Martínez-Cervera S, Fiszman SM (2008) Distinctive sensory features introduced by resistant starch in baked products. LWT Food Sci Technol 41:1927–1933. https://doi.org/10.1016/j.lwt.2008.01.012

Bello-Perez LA, Flores-Silva PC, Agama-Acevedo E, Tovar J (2020) Starch digestibility: past, present, and future. J Sci Food Agric 100:5009–5016. https://doi.org/10.1002/jsfa.8955

Codină GG, Franciuc SG, Mironeasa S (2016) Rheological characteristics and microstructure of milk yogurt as influenced by quinoa flour addition. J Food Qual 39:559–566. https://doi.org/10.1111/jfq.12210

Crispín-Isidro G, Lobato-Calleros C, Espinosa-Andrews H et al (2015) Effect of inulin and agave fructans addition on the rheological, microstructural and sensory properties of reduced-fat stirred yogurt. LWT Food Sci Technol 62:438–444. https://doi.org/10.1016/j.lwt.2014.06.042

Del Nobile MA, Chillo S, Mentana A, Baiano A (2007) Use of the generalized Maxwell model for describing the stress relaxation behavior of solid-like foods. J Food Eng 78:978–983. https://doi.org/10.1016/j.jfoodeng.2005.12.011

Dogan M, Kayacier A, Toker ÖS et al (2013) Steady, dynamic, creep, and recovery analysis of ice cream mixes added with different concentrations of xanthan gum. Food Bioprocess Technol 6:1420–1433. https://doi.org/10.1007/s11947-012-0872-z

Ferry JD (1980) Viscoelastic properties of polymers, 3rd edn. Wiley, New York

Flores-Mancha MA, Ruíz-Gutiérrez MG, Rentería-Monterrubio AL et al (2021) Stirred yogurt added with beetroot extracts as an antioxidant source: rheological, sensory, and physicochemical characteristics. J Food Process Preserv 45:10. https://doi.org/10.1111/jfpp.15628

Fuentes-Zaragoza E, Riquelme-Navarrete MJ, Sánchez-Zapata E, Pérez-Álvarez JA (2010) Resistant starch as functional ingredient: a review. Food Res Int 43:931–942. https://doi.org/10.1016/j.foodres.2010.02.004

Gustaw W, Kordowska-Wiater M, Kozioł J (2011) The influence of selected prebiotics on the growth of lactic acid bacteria for bio-yoghurt production. Acta Sci Pol Technol Aliment 10:455–466

He J, Han Y, Liu M et al (2019) Effect of 2 types of resistant starches on the quality of yogurt. J Dairy Sci 102:3956–3964. https://doi.org/10.3168/jds.2018-15562

Hoover R, Hughes T, Chung HJ, Liu Q (2010) Composition, molecular structure, properties, and modification of pulse starches: a review. Food Res Int 43:399–413. https://doi.org/10.1016/j.foodres.2009.09.001

Jia S, Zhao H, Tao H et al (2022) Influence of corn resistant starches type III on the rheology, structure, and viable counts of set yogurt. Int J Biol Macromol 203:10–18. https://doi.org/10.1016/j.ijbiomac.2022.01.027

Jiang Y, Zhao Y, Zhu Y et al (2019) Effect of dietary fiber-rich fractions on texture, thermal, water distribution, and gluten properties of frozen dough during storage. Food Chem 297:124902. https://doi.org/10.1016/j.foodchem.2019.05.176

Keogh MK, O'Kennedy BT (1998) Rheology of stirred yogurt as affected by added milk fat, protein and hydrocolloids. J Food Sci 63:108–112. https://doi.org/10.1111/j.1365-2621.1998.tb15687.x

Lamsal BP, Faubion JM (2009) The beneficial use of cereal and cereal components in probiotic foods. Food Rev Int 25:103–114. https://doi.org/10.1080/87559120802682573

Langkilde AM, Champ M, Andersson H (2002) Effects of high-resistant-starch banana flour (RS(2)) on in vitro fermentation and the small-bowel excretion of energy, nutrients, and sterols: an ileostomy study. Am J Clin Nutr 75:104–111. https://doi.org/10.1093/ajcn/75.1.104

Li R, Ding L, Guo K et al (2023) The effects of different types of high-amylose maize starches on viscosity and digestion of acidified milk gels. Food Chem 404:134525. https://doi.org/10.1016/j.foodchem.2022.134525

Mwizerwa H, Abong GO, Okoth MW et al (2017) Effect of resistant cassava starch on quality parameters and sensory attributes of yoghurt. Curr Res Nutr Food Sci 5:353–367. https://doi.org/10.12944/CRNFSJ.5.3.21

Noosuk P, Hill SE, Farhat IA et al (2005) Relationship between viscoelastic properties and starch structure in rice from Thailand. Starch Stärke 57:587–598. https://doi.org/10.1002/star.200500401

Rao MA (2014) Rheology of food gum and starch dispersions, Food Engin. Springer

Rayment P, Ross-Murphy SB, Ellis PR (1998) Rheological properties of guar galactomannan and rice starch mixtures II. Creep measurements. Carbohydr Polym 35:55–63. https://doi.org/10.1016/S0144-8617(97)00231-2

Rezaei R, Khomeiri M, Kashaninejad M et al (2017) Steady and dynamic rheological behaviour of frozen soy yogurt mix affected by resistant starch and β-glucan. Int J Food Prop 20:S2688–S2695. https://doi.org/10.1080/10942912.2017.1397692

Rezaei R, Khomeiri M, Kashaninejad M et al (2018) Steady and dynamic rheological behaviour of frozen soy yogurt mix affected by resistant starch and β-glucan. Int J Food Prop 20:S2688–S2695. https://doi.org/10.1080/10942912.2017.1397692

Sah BNP, Vasiljevic T, McKechnie S, Donkor ON (2016) Physicochemical, textural and rheological properties of probiotic yogurt fortified with fibre-rich pineapple peel powder during refrigerated storage. LWT Food Sci Technol 65:978–986. https://doi.org/10.1016/j.lwt.2015.09.027

Soler A, Mendez-Montealvo G, Velazquez-Castillo R et al (2020) Effect of crystalline and double helical structures on the resistant fraction of autoclaved corn starch with different amylose content. Starch Stärke 72:1900306. https://doi.org/10.1002/star.201900306

Szołtysik M, Kucharska AZ, Dąbrowska A et al (2021) Effect of two combined functional additives on yoghurt properties. Foods 10:1–15. https://doi.org/10.3390/foods10061159

Tejada-Ortigoza V, García-Cayuela T, Welti-Chanes J et al (2020) Emerging technologies for the extraction and modification of dietary fiber, Food Engin. Springer

Wang S, Li C, Copeland L et al (2015) Starch retrogradation: a comprehensive review. Compr Rev Food Sci Food Saf 14:568–585. https://doi.org/10.1111/1541-4337.12143

Wang C-C, Yang Z, Xing J-J et al (2021) Effects of insoluble dietary fiber and ferulic acid on the rheological properties of dough. Food

Hydrocoll 121:107008. https://doi.org/10.1016/j.foodhyd.2021.107008
Witczak M, Smółka Z, Witczak T et al (2020) Influence of replacement part of starch with inulin on the rheological properties of pastes and gels based on potato starch. Int J Food Sci 2020:7642041. https://doi.org/10.1155/2020/7642041
Zhong Q (2019) Chapter 18—Food rheology. In: MBT-H of F (ed) Kutz dairy and food machinery engineering, 3rd edn. Academic Press, pp 461–481