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Rheological and Textural Properties of Yogurts Enriched with Jerusalem Artichoke Flour

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Abstract

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Rheological and textural properties of whole milk yogurt enriched with the flour from tubers of two Jerusalem artichoke cultivars were investigated. They were compared with the properties of yogurts enriched with the extracts containing fructooligosaccharides and other carbohydrates isolated by different methods from tuber flour. Chemical analysis of extracts was also performed. All yogurts enriched with tuber flour or other carbohydrates exhibited viscoelastic behaviour.

Keywords: whole milk yogurt; supplementation; parameters; carbohydrates; chemical analysis

Yogurt is a popular milk product. To increase health promoting features, yogurt can be supplemented with prebiotic ingredients. Inulin and fructooligosaccharides (FOS) are the best known prebiotics whose beneficial influence on human health was shown many times. They improve intestinal microflora by stimulating the growth of beneficial bacteria in the large intestine and can inhibit bacterial and viral infections by modulating host defence responses. Inulin and FOS may help to counteract colorectal cancer and other diseases (Gibson *et al.* 2005; Di Bartolomeo *et al.* 2013). Moreover, inulin and FOS serve as dietary fibre, and reduce blood cholesterol and increase calcium absorption (Kaur & Gupta 2002).

Nowadays, chicory roots are the main source of inulin. Another plant of the family *Compositae*, Jerusalem artichoke (*Helianthus tuberosus* L.), also has a high biotechnological potential and is considered as a source of fructan-based oligosaccharides for industry. The studies on Jerusalem artichoke cultivation in different soil and climatic conditions, on the carbohydrate composition of tubers of various cultivars and their changes at different phases of vegetative growth and different conditions of storage after harvest have been carrying out (Matias *et al.* 2013; Krivorotova & Sereikaite 2014).

Usually, low-fat yogurts are supplemented with inulin considering it as a fat and sugar replacer. The influence of inulin on physicochemical and sensory properties of yogurts depends on the amount of inulin added to yogurt and on the degree of polymerisation (DP) of the inulin chain (Meyer *et al.* 2011). There are only a few publications on properties of whole milk yogurts supplemented with inulin or FOS (Guggisberg *et al.* 2009; Cruz *et al.* 2013). Jerusalem artichoke flour could be an ideal prebiotic material for dairy industry. The flour contains not only fructooligosaccharides, but also the natural plant cell wall that could serve as a dietary fibre comprising mixed polysaccharides: cellulose, hemicellulose and pectin (Harris & Smith 2006). The flour from Jerusalem artichoke tubers can be easily produced, and its application in sausage and bread has been

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studied (Praznik *et al.* 2002; Afoakwah *et al.* 2015). To our knowledge, there is a lack of information on the application of tuber flour for yogurt production and the influence on its properties.

Here, the rheological and textural properties of whole milk yogurt enriched with the flour from tubers of two Jerusalem artichoke cultivars Albik and Rubik are presented. They are compared with the properties of yogurts enriched with the extracts containing FOS and other carbohydrates isolated by different methods from tuber flour. Chemical analysis of extracts was also performed. For comparison reasons, the whole milk yogurts supplemented with sucrose or chicory inulin were also prepared.

MATERIAL AND METHODS

Two Jerusalem artichoke cultivars Albik and Rubik were organically grown in South Lithuania. Whole milk powder was purchased from a Polish producer. Freeze-dried yogurt thermophilic lactic cultures (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*) and inulin from chicory Frutafit® IQ were kindly delivered by Chr. Hansen Poland Sp. z.o.o. and Sensus B.V., respectively. Glucose, fructose, and sucrose from Sigma-Aldrich (USA) and 1-kestose (GF_2), nystose (GF_3) and 1^F-fructofuranosyl nystose (GF_4) from Wako (Japan) were applied.

Preparation of Jerusalem artichoke tuber flour. Fresh tubers were washed, peeled, cut into small pieces (about 0.5 cm of thickness), and blended. The homogenised plant material was dried at 80°C for 24 h and milled.

Preparation of FOS from Jerusalem artichoke tuber flour. *Method 1 (M1):* 10 g of flour was suspended in 200 ml of distilled water and heated in a water bath at 85°C for 30 min with gentle mixing several times. After cooling to 40°C the wet plant pulp was removed by centrifugation at 9000 *g* for 20 minutes. Then the extract was concentrated three times by evaporation at reduced pressure at 80°C. Finally, it was dried in the oven at 55°C for 3 days.

Method 2 (M2): The extract was prepared as described above, and after pulp removing it was additionally clarified with activated carbon. Activated carbon up to four grams was added and the samples were incubated at 60°C in water bath for 15 minutes. Then they were centrifuged at 9000 *g* for 20 min and, in addition, filtered through filter paper. Finally, the extract was concentrated and evaporated as described in Method 1.

Method 3 (M3): FOS were extracted from 10 g of flour suspended in 200 ml of water acidified with acetic acid to pH ~ 4. The following steps were the same as in Method 2.

Method 4 (M4): The extracts were obtained as described in Method 1 until the step of concentration at reduced pressure. Then ethanol or acetonitrile was added to 20% syrup (at 5 : 1, v/v). The sample was kept one hour at 5°C. Then the precipitate was centrifuged at 6000 *g* for 20 min and dried overnight at 50°C.

Determination of FOS and other carbohydrates by HPLC. Carbohydrate content in the flour extracts was analysed using a Viscotek chromatographic system equipped with a refractive index detector and Prevail Carbohydrate ES column (250 mm × 4.6 mm, 5 μm; Alltech, USA). Chromatographic process was performed at 30°C using the mixture of acetonitrile/water (70 : 30, v/v) as a mobile phase at a flow rate of 1.0 ml/ minute. Before injection, the solution of extract (12 mg/ml) was diluted with a mobile phase (3 : 2, v/v) and filtered through 0.2 μm filter. The percentage of fructose, glucose, sucrose, GF_2, GF_3, and GF_4 in the extracts (% w/w) was calculated using standard curves.

NMR spectroscopy. ^{1}H NMR measurements in D_2O were performed on Brucker 400 Ascend™ equipment at 400 MHz. The concentration of samples was equal to 0.1 mg/ml.

Cryoscopic measurements. For the determination of the average molecular weight of carbohydrates and degree of their polymerization, cryoscopic measurements were performed as described previously (Glibowski & Wasko 2008).

Yogurt preparation. The mixture of 12 g of whole milk powder and 2, 4, or 8 g of Jerusalem artichoke tuber flour extract or 4 g of tuber flour was dispersed in 75 ml of deionised water with moderate mixing using a magnetic stirrer. Then deionised water was added to the final sample weight of 99 g. All samples were heated in water bath at 80°C for 30 min and subsequently cooled down to the fermentation temperature. Then 1 ml of 0.77% previously prepared suspension of yogurt culture was added, and the samples in plastic containers with twist lids were incubated at 45°C for 5 h in a thermostatic cabinet. Then yogurts were stored at the temperature of 5°C. All samples were prepared in duplicate and used for further analysis. Each sample was analysed in

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triplicate. For comparison reasons, the whole milk yogurts supplemented with sucrose or Frutafit® IQ chicory inulin (the average DP ≥ 10, producer's data) were also prepared. The pH of samples was measured using a CP-401 pH meter (Elmetron Sp.J., Poland).

Rheological measurements. Rheological measurements were conducted using a Haake RS 300 rheometer with parallel-plate geometry (35 mm diameter). When the sample was placed on the plate, the lift moved and the plate took the measuring position (1 mm gap between parallel plates). All measurements were carried out at 21°C. The apparent viscosity was measured at a shear rate of 20 s^{-1} for 120 s, as previously measured (Glibowski & Kowalska 2012). All rheological data were collected and calculated by Haake Rheowin software version 3.61.0004.

Texture analysis. The penetration test was performed by driving a cylindrical probe (1 cm diameter) with the crosshead speed of 1 mm/s into a 15-mm depth, using a TA-XT2i texture analyser. The maximum peak value after the probe was driven 15 mm into the sample was considered as yogurt hardness (g). The analysis was performed without removing the samples from the containers.

RESULTS AND DISCUSSION

For the isolation of FOS and other carbohydrates, four methods were applied. The highest yield of dry mass of extracts (76–81%) was obtained by M3 and the lowest one by the precipitation with ethanol or acetonitrile (10–13%). The carbohydrates composition of extracts was analysed by HPLC (Table 1 and Figure 1). In the chromatogram of the extract obtained by M3 new peaks appear with a retention volume of 10.6, 14.7, and 20.2 ml. It is plausible that these peaks correspond to inulobiose, inulotriose, and inulotetraose (Mutanda *et al.* 2008). All the extracts contain FOS with DP > 4 (Figure 1). The lowest average molecular weight (MW) and degree of polymerisation (DP) calculated by a cryoscopy method were determined for extracts obtained by M3. The highest ones were determined for carbohydrates precipitated with ethanol (Table 2).

In addition, ^{1}H NMR spectra of carbohydrates obtained by four methods were recorded (Figure 2). The extracts obtained by M1, M2, and M3 contain some organic and amino acids (Figures 2A–C). In the case of M4, malic acid and unknown compound (1.15 ppm, t) were found in the ^{1}H NMR spectrum (Figure 2D) (Bertram *et al.* 2010). In the region of 3.03–4.3 ppm at the middle frequency of magnetic field, there are proton signals from various carbohydrates including free fructose, free glucose, sucrose, and fructooligosaccharides (Figure 2).

DP of carbohydrates was also calculated using NMR spectroscopy (Table 2). The determination was based on the assumption that each chain of fructooligosaccharides contains one molecule of glucose. DP was calculated from the ratio of the proton integration value of the H-1 signals of terminated glucose at 5.44 ppm to that of the H-3 (or H-4) signals of fructosyl residues at 4.27 (or 4.11) ppm (Cérantola *et al.* 2004).

Prior to rheological measurements, the acidification of yogurts was measured (Table 3). The pH values of yogurts supplemented with Albik or Rubik flour were similar to those of yogurts with sucrose or inulin IQ. However, the pH values of yogurts supplemented with carbohydrates differed. The changes in pH of some yogurts were analysed after storage

Table 1. The percentage of carbohydrates in the extracts obtained by different methods

Carbohydrates	Albik					Rubik			
	method of isolation								
	1	2	3	4^a	4^b	1	2	3	4^a
Fructose	1.2	1.3	19.1	6.4	0.7	0.8	2.1	21.1	0.9
Glucose	0.2	0.1	1.9	0.8	0.1	0.0	0.5	2.1	0.0
Sucrose	8.1	6.9	14.9	7.5	6.1	1.3	4.0	13.5	17.8
GF_2^c	6.0	5.3	6.6	3.3	4.9	0.8	3.4	3.7	10.9
GF_3^c	5.2	4.2	5.1	2.8	4.3	2.6	3.2	3.4	7.7
GF_4^c	5.2	4.4	5.8	5.2	4.2	4.8	2.8	0.8	7.6

[a]ethanol; [b]acetonitrile were used as organic solvents; [c]GF_2, GF_3, and GF_4 correspond to 1-kestose, nystose, and 1^F-fructofuranosyl nystose, respectively

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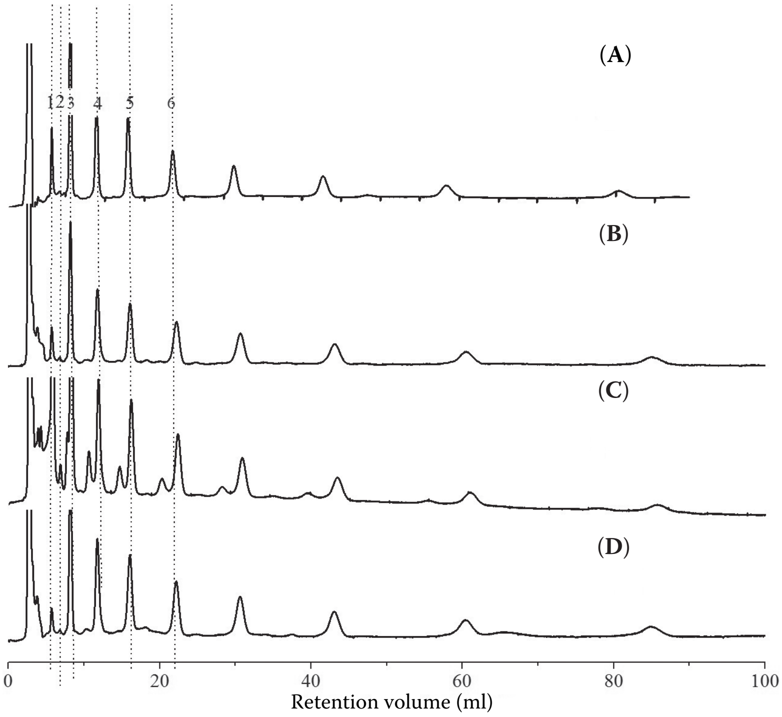

Figure 1. HPLC analysis of FOS and other carbohydrates extracted from Albik tuber flour by M1 (**A**), M2 (**B**), M3 (**C**), and M4 (**D**, precipitation with acetonitrile)

The peaks 1, 2, 3, 4, 5, and 6 correspond to fructose, glucose, sucrose, kestose, nystose, and 1F-fructofuranosyl nystose, respectively

at 5°C for 20 days. Post-acidification of yogurts was negligible (Table 3).

In our study, yogurts were analysed by means of the small strain deformation technique. In all cases, the yogurts exhibited higher G' than G'' values (Figure 3 and Table 3). It means that yogurts showed more elastic than viscous behaviour and were characterised as weak gels (Meyer *et al.* 2011). The storage (G') and loss (G'') modulus was slightly dependent on the frequency at its low range. For all yogurts, tan δ is < 1 and points to dominant elastic properties. Various supplements influenced the firmness of the gel in a different way. Yogurts supplemented with the extract of carbohydrates (M3 for both tuber cultivars) show similar firmness as compared with yogurts supplemented with sucrose. The carbohydrates isolated by M3 have the lowest average MW and the high percentage of fructose and sucrose (Tables 1 and 2) and, therefore, the yogurt has similar properties like the yogurt containing sucrose. The yogurts containing carbohydrates extracted by M2 and M4 (an average DP is equal approximately to 3 and 5, respectively) are softer (G' values are approximately 2.5–4.5 fold lower than the G' value of yogurt containing sucrose). These data are in accordance with earlier studies. Cruz *et al.* (2013) found that the plain yogurt supplemented with the oligofructose Raftilose P95 exhibited lower G´ values than the yogurt without oligosaccharides. The average DP of Raftilose P95 is equal to 4 and similar to DP of our carbohydrates obtained by M2 and M4. However, the whole milk yogurts containing inulin IQ are stiffer (G' value is approximately 2-fold higher than G' value of yogurt containing sucrose). The decrease of G' and G'' values of yogurts supplemented with inulin or carbohydrates isolated by M2 from Rubik flour

Table 2. The average molecular weight of carbohydrates and their degree of polymerisation[a]

Parameter	Albik				Rubik			
	method of isolation							
	1	2	3	4[c]	1	2	3	4[b]
MW[b]	490 ± 2	430 ± 1	330 ± 1	830 ± 25	370 ± 1	380 ± 1	240 ± 2	550 ± 2
DP	3.0	2.6	2.0	5.1	2.3	2.3	1.5	3.4
DP[NMR]	3.0	2.8	–	5.1	3.4	3.3	–	–

[a]molecular weight and degree of polymerisation correspond to MW and DP, respectively; [b]data are presented as mean ± standard deviation of four parallel experiments; [c]ethanol was used as an organic solvent

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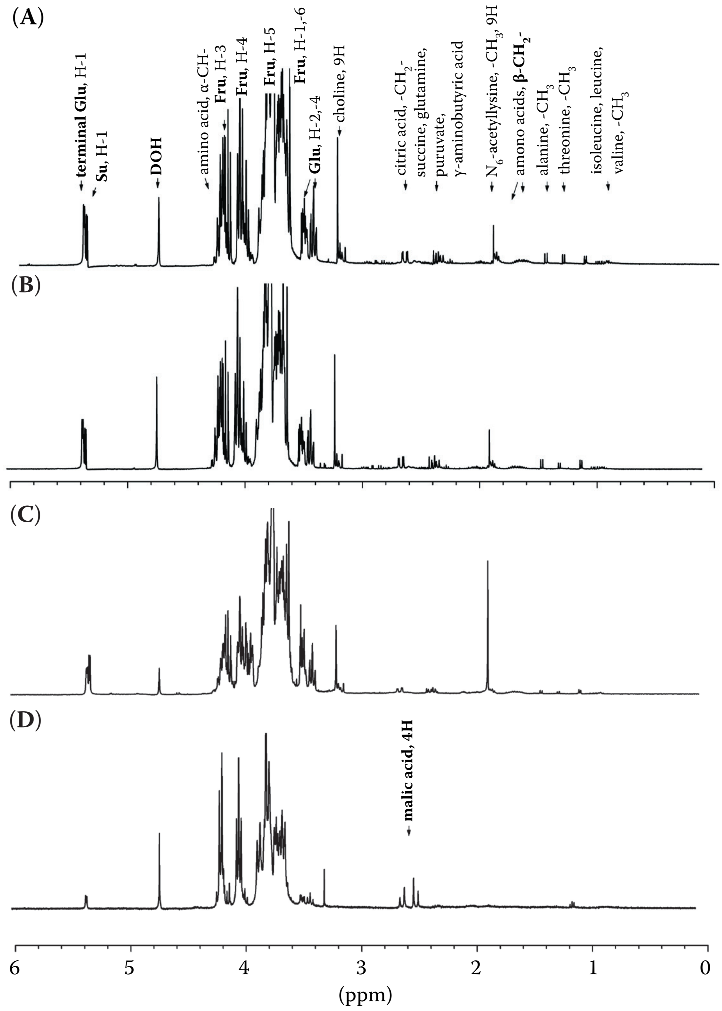

Figure 2. ^{1}H NMR spectra of extracts obtained from Albik tuber flour by M1 (**A**), M2 (**B**), M3 (**C**), and M4 (**D**, precipitation with acetonitrile)

Table 3. Viscoelastic parameters[a] and pH values of yogurts prepared with 4% of supplements[b,c]

Supplement	G' (Pa)	G'' (Pa)	tan δ	η (Pa·s)	pH	
					5°C, 18 h	5°C, 20 days
Sucrose	540 ± 37	170 ± 21	0.31	–	4.32 ± 0.01	–
Inulin IQ	1290 ± 140	380 ± 34	0.29	–	4.39 ± 0.04	4.28 ± 0.06
Albik flour	170 ± 20	48 ± 7	0.28	2.8 ± 0.3	4.37 ± 0.04	–
AF_2	200 ± 32	54 ± 7	0.27	3.2 ± 0.5	4.63 ± 0.04	4.57 ± 0.04
	280 ± 9[d]	107 ± 20[d]	0.38[d]	4.7 ± 0.3[d]	4.47 ± 0.02[d]	–
AF_3	460 ± 70	130 ± 44	0.28	7.5 ± 0.1	4.81 ± 0.04	–
	401 ± 63[e]	111 ± 45[e]	0.28[e]	6.6 ± 0.5[e]	4.80 ± 0.02[e]	–
AF_4	200 ± 53	46 ± 14	0.23	–	4.53 ± 0.05	4.48 ± 0.01
Rubik flour	1100 ± 160	370 ± 50	0.34	18.5 ± 3	4.44 ± 0.04	–
RF_2	140 ± 46	41 ± 10	0.29	2.4 ± 0.7	4.64 ± 0.03	4.64 ± 0.01
	450 ± 38[d]	140 ± 4[d]	0.31[d]	7.5 ± 2.1[d]	4.50 ± 0.01[d]	–
RF_3	290 ± 70	101 ± 23	0.35	4.8 ± 2.1	4.82 ± 0.06	–
	280 ± 56[e]	90 ± 38[e]	0.31[e]	4.8 ± 1.9[e]	4.86 ± 0.03[e]	–
RF_4	120 ± 54	34 ± 18	0.28	–	4.86 ± 0.03	4.77 ± 0.01

[a]viscoelastic parameters were determined at a frequency of 10 Hz; [b]carbohydrates extracted from Albik (AF_2, AF_3, AF_4) and Rubik (RF_2, RF_3, RF_4) flour by M2, M3, and M4, respectively; [c]data are presented as mean ± standard deviation of six parallel experiments; [d,e]supplement concentration was equal to 2 and 8%, respectively

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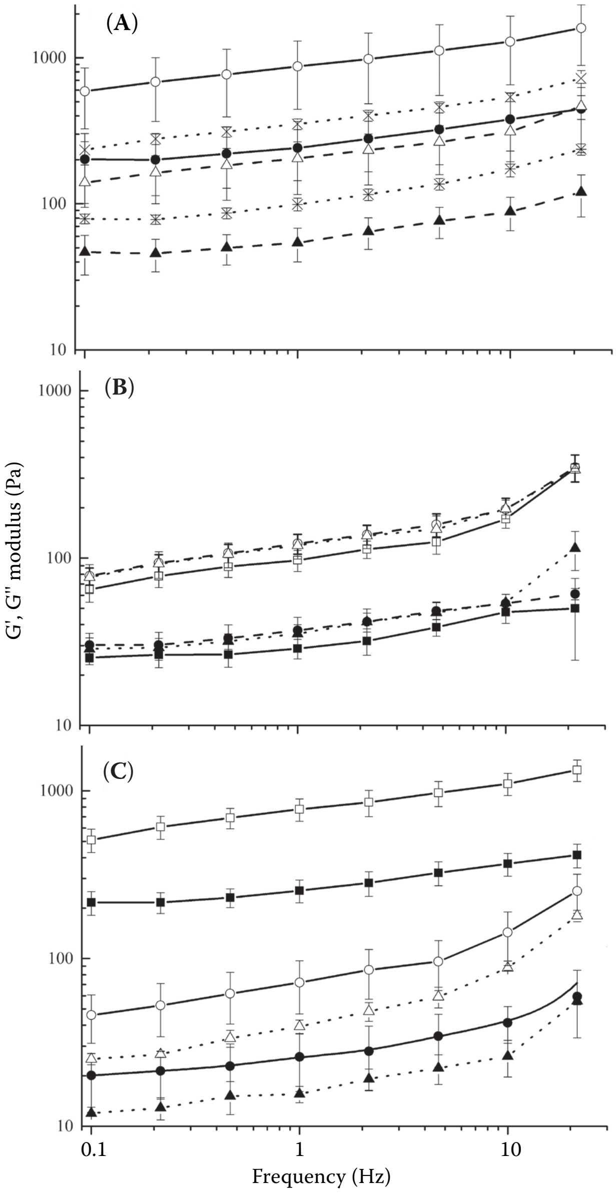

Figure 3. Mechanical spectrum of yogurts enriched with 4% of supplement: (**A**): sucrose (×, ✳), 18 h of yogurt storage; inulin Frutafit® IQ, 18 h (○, ●) and 20 days of yogurt storage (Δ, ▲); (**B**): Albik flour (□, ■), 18 h of yogurt storage; Albik carbohydrates isolated by M2, 18 h (○, ●) and 20 days of yogurt storage (Δ, ▲); (**C**): Rubik flour (□, ■), 18 h of yogurt storage; Rubik carbohydrates isolated by M2, 18 h (○, ●) and 20 days of yogurt storage (Δ, ▲); G' (○,×, Δ, □), G'' (●, ✳, ▲, ■)

was observed after storage for 20 days (Figure 3). This effect was negligible for yogurts enriched with carbohydrates from Albik flour. Therefore, the inulin of the average DP ≥ 10 influences the formation of casein and whey protein network and increases the rigidity of yogurt. Inulin as an amphiphilic molecule can form many hydrogen bonds with protein molecules and increases the networking of gel (Kip *et al.* 2006). A similar effect was obtained using Rubik flour. The gels were stiffer as compared with yogurts enriched with sucrose. However, the yogurts supplemented with Albik flour were softer (G' was equal to 170 ± 20) (Table 3) and similar to those supplemented with carbohydrates (M2). Moreover, yogurts enriched with Rubik flour had a high apparent viscosity as compared with the Albik flour supplement as well as the extracts of carbohydrates obtained by four different methods. The literature data are contradictory. Carrot cell wall particles (McCann *et al.* 2011) as well as orange (Sendra *et al.* 2010) and asparagus (Sanz *et al.* 2008) cell wall fibre increased the rigidity of yogurts. However, Garcia-Perez *et al.* (2006) reported that orange cell wall fibre had even a negative effect on gel strength.

The data of the penetration test is summarised in Table 4. The addition of carbohydrates extracted by M3 had no influence on yogurt hardness as compared with sucrose. However, a decrease of hardness was observed for yogurts enriched with carbohydrates extracted by M2. The decrease was significant especially for the extracts from tubers of Rubik cultivar. It is plausible that the changes of yogurt hardness are related to higher average MW of carbohydrates (Table 2) as compared with sucrose and the carbohydrates extracted by M2. The obvious changes were observed for yogurts supplemented with Albik or Rubik flour. The hardness of yogurts decreased more

Table 4. Hardness of yogurts prepared with 4% of supplements[a,b]

Supplement	Hardness (g)
Sucrose	18.2 ± 1.1
Inulin IQ	19.1 ± 0.4
Albik flour	6.8 ± 0.7
AF_2	12.8 ± 2.2
AF_3	14.8 ± 0.3[c] 19.0 ± 1.2 15.0 ± 1.3[d]
Rubik flour	8.2 ± 2.8
RF_2	7.0 ± 0.1
RF_3	15.1 ± 0.2[c] 18.2 ± 0.5

[a]carbohydrates extracted from Albik (AF_2, AF_3) and Rubik (RF_2, RF_3) flour by M2 and M3, respectively; [b]mean ± standard deviation of six parallel experiments; [c,d]supplement concentration was equal to 2 and 8%, respectively

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than twice. It is plausible that these changes occur due to the high degree of system heterogeneity. The size of flour particles cannot be comparable with the parameters of protein or inulin molecules. Flour particles incorporated into the yogurt gel undergo swelling and modify hydrogen bonding between water and milk protein molecules inducing rearrangements of the gel network and affecting textural properties. Moreover, the flour particle is a complex biopolymer composed of non-ionic and ionic molecules at the appropriate pH values. Recently, it has been shown that polysaccharides with different ionic charge influence the textural properties of acid milk gels differently (Pang *et al*. 2015).

CONCLUSIONS

All yogurts supplemented with Albik or Rubik flour or other carbohydrates exhibited viscoelastic behaviour. However, the rheological and textural properties of yogurt depended on carbohydrate composition and on the method of their isolation from tuber flour. To our knowledge, this is the first time when the tuber flour application in yogurts is described in detail. The flour of Jerusalem artichoke tubers could be used for yogurt production because inulin isolation from tubers is an expensive and time-consuming process. Further studies concerning flour milling characteristics and the effect of particle size on the rheological and textural properties of yogurt should be done.

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