Application of new insoluble dietary fibers from triticale as supplement in yoghurt - effects on physicochemical, rheological and quality properties

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ABSTRACT

BACKGROUND: The need to increase the daily intake of dietary fibers opens a new chapter in the research of functional foods enriched with fibers. Potential application of innovative product - insoluble dietary fibers from triticale in yoghurts, deployed by characterizing their food application and evaluating physicochemical, rheological and sensory properties was the aim of this research.

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RESULTS: Detailed characterizations of these fibers are presented for the first time and showed very good hydration properties, optimal pH (slightly acidic), optimal chemical composition, high antioxidant capacity which was proven by phenolics contents. Besides, these fibers showed negligible calorific value, with no phytates and high antioxidant capacity, mainly from ferulic acid. Therefore they could be successfully added to yoghurt. Enrichment of yoghurt having different milk fat content (1.5 and 2.8% w/w) with triticale insoluble fiber (1.5 and 3.0% w/w) significantly influenced the syneresis level, its apparent viscosity, yield stress and thixotropic behavior. The overall sensory quality scores indicated that yoghurt enriched with 1.5% triticale insoluble fibers was recognized as 'excellent' and had enhanced antioxidant activity.

CONCLUSIONS: Insoluble triticale fiber could therefore be used as a supplement to produce functional yoghurt.

Key words: yoghurt, triticale, insoluble dietary fiber, rheology, antioxidant activity

INTRODUCTION

The International Life Science Institute established that "a food product can be considered as functional if it has satisfactorily been proved that it produces a beneficial effect on one or more physiological functions, besides its conventional nutritional effects, being this relevant for improving the human health and/or reducing the risk of suffering certain diseases"[1]. Insoluble dietary fibers (IDF) is known to increase stool weight and decrease colonic transit time[2], which ensures prevention of colonic diverticulosis and constipation[3]. The second important benefit of IDF is their antioxidant capacity that comes from phenolic and may contribute with their health benefits[4]. Despite the proven healthful influence of IDF on chronic disease risk reduction, the average worldwide ingestion of this component is considered as low. There is pressing need for increasing of daily intake of IDF.

Triticale IDF (IDFT) is innovative product [5] and can be considered as a good candidate for the addition in functional foods. IDFT are low-calorie, non-allergenic dietary fibers obtained by innovative technology from triticale (x *Triticosecale*, Wittmak) – hybrid of wheat (*Triticum* sp.) and rye (*Secale* sp.). Triticale contains high levels of nutritionally beneficial compounds such as phenolic acids and IDF[6, 7]. Its cultivated in more than 30 countries [8] and has many benefits compared to other crops, but insufficiently utilized as yet[9]. Triticale is unjustified very little represented in the human diet [10]. Usage of IDFT in some dietary products could increase the contribution of triticale in human diets, daily intake of IDF and cultivation of this interesting grain.

Fermented dairy product already have a positive health image due to the beneficial action of its viable bacteria[11]. Yoghurt is a very popular fermented dairy product and represents a good base for development of enriched food in order to improve its nutritional value and health benefits. In this sense it has been shown that it is possible to enrich the yoghurt with IDF from a number of sources – soy, rice, oat, corn and sugar beet [12]. Use of new sources of fiber is recommended for several reasons: (a) to develop novel food considering technological and physiological functionality of DF depending on its sources and chemical composition; (b) to contribute to the social and economic development of local producers by successfully applying the fibers of native plants in food; and (c) to promote higher acceptability of fiber-rich products, using local and traditionally consumed plants as sources of DF [13]. The main criteria for the acceptability of fiber-enriched foods are: good processing behavior; good stability and appearance and satisfactory sensory properties [14].

The aim of this study was to investigate potential application of IDFT in yoghurt production. Hence, the objective of the present work is twofold: (1) to characterize IDFT and identify possibilities for food application and (2) to evaluate and compare the composition, syneresis, and rheological behavior, textural and sensory properties of stirred yoghurt having different fat and insoluble fiber contents. Results should provide important insights in the potential commercialization of this fiber as a novel functional component of dairy products.

MATERIALS AND METHODS

Characterization of triticale fibers

IDFT were obtained by innovative environmental friendly technology [5] based on auto hydrolytic properties of triticale. Triticale has its own α-amylases which can hydrolyze all starch present in grain, which was already used for bioethanol production[15] and for fungal amylase production[16]. This characteristic was used in the IDFT production technology, too. IDFT are without taste and odor, low-calorie and non-allergenic.

Elemental microanalyses were performed on a elemental analyzer (LecoCHNS-932, LECO Corporation, USA), by the standard micromethod. Elemental concentrations of trace elements were determinate by ICP – AES (Inductively Coupled Plasma Atomic Emission Spectroscopy).

Hydration properties of DF are described by three commonly used characteristics – the swelling capacity (SC) – water and fat, water retention capacity (WRC) and fat/oil retention capacity (ORC). These characteristics were determined on 25 °C according to standardized procedures [17].

Specific hydration properties of IDFT were determinate in accordance with the requirements of the yoghurt preparation, because it was added in yoghurt during preparation process. In this way, IDFT suffered temperature changes following the yoghurt process, and it is known that the hydration properties of the fibers depend on the temperature and the incubation time. We have also introduced a new specific feature of the fiber – milk retention capacity (MRC) because IDFT come into contact with milk during the yoghurt process, and milk and water are not the same, and the same behavior of fibers in terms of swelling and retention is not expected. WRC, MRC and ORC were determinate after incubation and centrifugation (1 min on 14,000 *g*) of IDFT in water, milk and butter respectively on 90 °C for 30 min, than cooled on 45 °C for 4 h.

pH of IDFT was determinate in 10% (w/V) IDFT suspension. The acidity was determined by titration with NaOH (0.1 mol/L) to pH 8.10, and the results were expressed as g citric acid/100 mL sample [18].

Moisture content analysis was done by determining content of water by drying method. The ash content analysis was performed by the method of incineration according to local standardized analysis methods for food industry.

IDFT were analyzed for contents of proteins in buffer extracts according to Bradford method [19], residual sugars were detected by 3,5-dinitrosalicylic acid (DNS) method [20], while eventual non hydrolyzed starch content were determinate using Iodine reagents [21]. Phytates content in IDFT were determinate according to color method for phytase activity [22].

Phenolic contents of IDFT were determinate in three extracts. Free and bound phenolics were extracted according to the procedures used for phenolics analysis in triticale bran [7]. Total phenolic content was determinate by Folin Ciocalteu method[23], expressed as ferulic acid equivalents (mg/100 g) based on triplicate measurements.

Individual phenolics concentrations (*p*-coumaric, ferulic, vanilic and syring acids) were analyzed by HPLC-DAD (Thermo Dionex Ultimate 3000 HPLC system,Thermo Dionex Cromeleon 7 software, USA). The separation of phenolics was accomplished on a reversed-phase Hypersil GOLD aQ-C18 column (150 x 3.0 mm). The mobile phases were A (TFA) and B (acetonitrile) at a flow rate of 0.5 mL/min, the injection volume was 20 μL for each sample. A gradient system was used as follows: 0 min 5% B, 0–29 min gradient 5-60% B; 30-37 min 5% B, 37-50 min 5% B. Elution of compounds of interest was monitored at a wavelength of 280 nm. For phenolic acid content, ferulic acid, *p*-coumaric, syring and vanilic acids in methanol was used as the standards and peaks were quantified based on areas and calculated as equivalents (μg/g IDFT).

Yoghurt production

Pasteurized and homogenized milk with different fat content (1.5 and 2.8g/100g, sample A and B, respectively) was used for yoghurt preparation. Yoghurts were prepared in three variants: control samples without fiber (A0 and B0), with 1.5% (1.5g IDFT/100g yoghurt, A1 and B1) and 3% (3g IDFT/100g, A2 and B2). Fibers were added before milk heat treatment. Starter culture ("Yoflex 812", Chr Hansen, Nieuwegein, Netherlands) was added in the amount of 0.02% (w/w). Fermentation was set at 43 °C until pH 4.6 was reached. Yoghurt samples were mixed and cooled during 24 h at temperature of 4-7 °C and analyzed.

Yoghurt characterization

The total solids, fat and proteins content in the yoghurts were determined by standard methods [24].

Rheological properties of stirred yoghurt samples were performed at 5 °C on a Rheometer (Kinexus Pro+, Malvern, United Kingdom) with the four blade vane as a tool. Data processing was performed using a software package, (Kinexus Version 1.60, Malvern Instruments Ltd., UK).

Flow behavior of yoghurt samples was characterized by thixotropic test. Shear stress was recorded at increasing and decreasing shear rates from 0 to 40/s within 50s (upward and downward flow curves). Thixotropic behavior of the samples was investigated by hysteresis loop area (A, Pa/s) which was determined as difference in area under upward and downward flow curves by a software (Origin 8.0, OriginLab Corp, USA). Apparent viscosity (η_{app}) was calculated at 10/s of shear rate.

Dynamic oscillation test was subsequently conducted following the flow behavior assessment to characterize the viscoelastic properties of yoghurt samples.

Strain sweeps were performed (0.001 – 500%) to determine the linear viscoelastic range (LVR) of samples at a constant frequency of 1 Hz. The yield stress value were calculated from the cross-over (G'=G'') point outside of the LVE [25].

Syneresis properties were analyzed by measurement of clear supernatant obtained after centrifugation (222 g/10 min) of 40 g of yoghurts at 4 ± 1 °C [26].

Texture properties of yoghurts such as firmness, consistency, cohesiveness and index of viscosity, were determined by a texture analyzer equipment (TA.HD, Stable Micro system, United Kingdom) through a single compression test, using a back extrusion cell (A/BE) disc and an extension bar, using 5 kg load cell at 5 °C. Tests were carried out in a standard size back extrusion container (50 mm diameter) approximately 75% full immediately after removal from storage.

Sensory quality rating was conducted by a sensory panel that consisted of 10 members experienced in dairy products quality judging. Quality grading was performed using 5-level quality scoring method. Overall sensory quality was assessed by evaluating four initially selected characteristics: appearance, odor, oral texture and flavor that were assigned appropriate values of coefficients of importance (CI): 3, 2, 9, and 6, respectively. In order to calculate the overall quality score, individual scores given to the selected sensory characteristics were first multiplied by the corresponding coefficients of importance, and then the sum of corrected score-values was divided by the sum of coefficients of importance.

All experimental samples were evaluated by the sensory panel in three replicates after both 2 and 9 d of storage at 4 °C after the yoghurt was manufactured.

Antioxidant capacity of IDFT and yoghurt enriched with IDFT

Antioxidant capacity of yoghurt, IDFT and yoghurt enriched with IDFT were determinate. For analysis, yoghurt with 2.8% milk fat and 1.5% IDFT was chosen as the best rated showed in the previous analysis.

ABTS radical scavenging assay

ABTS radical cation (ABTS•+) was produced by reacting 7 mmol/L ABTS stock solution with 2.45 mmol/L ammonium persulfate and allowing the mixture to stand in the dark at room temperature for 12–16 h before use. The ABTS•+ solution was diluted with methanol to an absorbance of 0.70±0.02 at 700 nm. Diluted ABTS•+ solution (3.9 mL) were mixed with 100 μL of sample or Vitamin C standard and reaction (decrease in absorbance) were monitored during 15 min. Vitamin C solutions of known concentrations were used for calibration. Antioxidant capacities were expressed as μmol Vitamin C/g of sample. ABTS radical scavenging assay was determinated also in all three phenolics fractions – extracts of IDFT bound, free and water dissolved).

Radical scavenging activity (RSA%) assay

RSA was measured according to Brand-William method[27]. An aliquot of sample (200 μL) was mixed with 2.9 mL of 60 μM DPPH (2,2-diphenyl-1-picrylhydrazyl) in methanol solution are vortexed. Reaction was carried out in the dark for 30 min. Antioxidant activity was expressed as percentage inhibition of the DPPH radical and was determinate by equation:

$$RSA\,\% = \frac{(Acontrol \quad Asample)}{A\ control}\ x\ 100$$

Determination of total phenolic content (TPC)

Total phenolic content was determinate by Folin Ciocalteu method[23], expressed as ferulic acid equivalents (mg/100 g) based on triplicate measurements.

Statistical analysis

Results of the effects of fat and fiber content on the yoghurts properties were analyzed using analysis of variance (ANOVA). The mean comparisons of the parameters were performed with post-hoc LSD test at $p < 0.05$.

The sensory data were first analyzed separately by 3-way ANOVA that included ‘samples’ as fixed factor, and ‘assessors’ and ‘replications’ as random factors. The model included main effects and all 2-way interactions. Tukey HSD test was used to separate the mean quality scores. In the second iteration, original sensory data were subjected to 4 way ANOVA that included ‘milk fat content’, ‘fiber content’, and ‘storage’ as fixed factors and ‘assessors’ as random factor. The model included main effects, all 2 way interactions, and the fixed factors 3 way interaction. The level of statistical significance was set at 0.01.

RESULTS AND DISCUSSION

Characterization of IDFT

Chemical analysis of IDFT, Table 1A and Table 1B, was intended to show the amounts of digestible components of IDFT – proteins, starch and sugar. All examined components were present in traces comparing to analogue cereal IDF [28]. These results confirm negligible caloric value and hypoallergenic properties of IDFT. Addition of IDFT in yoghurt will not increase caloric value. This is important for functional foods enriched with IDF that could be part of a menu of people on restrictive diets.

There were no phytates in IDFT, Table 1A. Consumption of food enriched by IDFT contributes to meeting the daily intake of IDF without negative consequences that arise by entering cereal or whole grains. Phytates from cereals associate with IDF, complexed with Zn^{2+}, Ca^{2+} and Fe^{2+} in digestive tract disables their resorption[29].

Elemental microanalysis of IDF is not common analysis in literature. Usually its take only one or few elements of interest [30]. It is known that triticale is rich in major mineral elements (K, P, Mg) and nutritionally important minor elements (Na, Mn, Fe, Cu, Zn) [31]. We showed that IDFT retain essential elements (Na, K, Fe, Mn, Mg, Ca, Co) from triticale, while there were no heavy metals, Table 1C.

Hydration properties of IDF contribute to stabilization of the structure of foods (dispersions, emulsions and foams) by modifying the rheological properties of the continuous phase and influence food texture. Capacity for fat absorption enhances their retention of fat that is normally lost during cooking, which could be beneficial for flavor retention and to increase the technological yield [14]. SC, WRC and ORC of IDFT presented in Table 2 are in range or higher than analogue IDF from cereals [28, 32].

Adding of IDF in food usually requires adjusting recipes for product preparation and/or special pretreatment of IDF because of their hydration properties[33]. In this respect we implemented the specific characterization of IDFT additionally, included the WRC, ORC and MRC on temperature and time correspond to yoghurt preparation process. The Specific WRC and ORC were higher than standard WRC and ORC on room temperature (Table 2A). It implies that usage of IDFT in yoghurt was a good target choice. Higher WRC, ORC and MRC indicate that IDFT could contribute to higher stability and to retention of original taste of yoghurt[33]. It also implies that specific characterization of IDF could be a good indicator in terms of behavior prediction of IDF during food preparation processes.

Table 2A confirms that pH, moisture and ash content fit within legal referent values. Slightly acidic pH of IDFT fit well with yoghurt preparation process which requires slightly acid environment. High acidity of IDFT, compared with other DF [18], will contribute to buffer capacity during yoghurt preparation process.

Antioxidant DF can be defined as a product containing significant amounts of natural antioxidants associated with the fiber matrix [34]. IDFT has very high antioxidant capacity, measurement in every phenolic fraction (Table 2B) and hence may be considered as a good candidate for antioxidant DF.

Comparisons of antioxidant capacities of IDF from cereals are very difficult due to the usage of different antioxidant tests and different expression pattern of results in literature. Nevertheless, analogue triticale fractions of phenolics showed similar relation in values of antioxidant capacity between bound and free phenolics [7].

Phenolics content of IDFT with HPLC analysis of individual components IDFT retained about 30% of total polyphenols relative to triticale bran. Most of phenolics were bounded in IDFT, as shown in triticale bran too [7]. In triticale the predominant phenolic acid is ferulic acid as well as in other cereals, where it serves as a bridge between lignin and polysaccharides (arabionxylans) via ether and ester bonds [35]. Other most common phenolic acids in triticale are p-coumaric and vanilin [7]. From results reported here it can be concluded that IDFT retained about 30% of ferulic acid and 100% of p-coumaric and vanilic acids from triticale as started material. Syringic acid was completely lost in the process of obtaining IDFT from triticale [7].

The characteristics of IDFT imply that this innovative product could be successfully added to a commonly used food without significantly changing the purpose and taste of them and at the same time making them functional foods.

Yoghurt characterization

Chemical composition and syneresis of yoghurts

Fermentation for all samples lasted about 4 h and pH values were not significantly influenced by addition of IDFT one day after production (data not presented). The yoghurts dry matter contents were influenced by different fat and IDFT contents while the significant influence ($p < 0.05$) on the protein content was not found (Table 3) which is important from allergenic point of view.

The fat and IDFT contents significantly influences ($p < 0.05$) on the syneresis level. As expected, the higher level of syneresis was found in yoghurts with lower fat content. Low fat products due to defects are usually produced with the addition of some ingredients in order to improve the textural properties [36]. Presence of insoluble fiber, such as IDFT, that does not incorporate within protein network contributes to a higher level of syneresis. Zare at al.[37] found that syneresis in 1–2% lentil flour supplemented yoghurts was significantly higher than all other samples. Opposite to that, other authors[38] showed that whey separation is reduced in yoghurts made with passion fruit fibers probably due to pectin present in this kind of fiber. Maximum concentration of triticale bran which could be acceptable for consumer perspective and stability of product is 4% [6] which was confirmed for IDFT in our research.

Rheological properties of yoghurts

Yoghurts exhibit a variety of non-Newtonian characteristics, such as viscoelasticity, time-dependency, yield stress and shear-thinning behavior [39].

A decrease in apparent viscosity (η_{app}) with increasing shear rate was found in all yoghurts indicating shear thinning behavior (Figure 1). Two factorial ANOVA showed that both factors, fat and IDFT, significantly influenced η_{app} value. The higher fat content in yoghurt contributed to the higher apparent viscosity, while addition of IDFT resulted in the decreasing of η_{app} ($p<0.05$). It has been confirmed that type of fiber influence on the apparent viscosity of yoghurts [40]. The structure of initial gel network and the breakdown process used to convert the gel into the stirred yoghurt affected the physical and sensory properties of products. Weak gels due to poor protein-protein interactions resulted in decreased stiffness of initial gels. This also contributed to decreased η_{app} and sensory properties of stirred yoghurts. The process parameters, such as preheating temperature and incubation temperature highly influence on the initial gel properties as well as on the final product [39].

Yoghurts show thixotropic behavior in which when a sample is sheared at increasing and then at decreasing shear rates, the hysteresis area between the curves is observed (Figure 2). Hysteresis loop area (A, Pa/s) presents the energy that is needed to destroy the structure of material. Our data showed that fat content significantly influenced on the hysteresis loop area ($p < 0.05$), while the effect of IDFT addition was not found. Yoghurts with the higher fat content require the higher energy to breakdown the structure.

The significant influence of fat and IDFT content on the yield stress was found ($p<0.05$) (Table 3). The yield stress value indicates the difficulty in breaking strands representing fracture properties of gel depends on the number of bonds per cross section of the strand as well as strength of each bond [41]. It also presents the minimum stress required to initiate flow [42]. The low value of yield stress in yoghurts with IDFT can be explained by the decreased possibility of interactions between protein aggregates due to presence of fiber in continuous phase surrounding the aggregates. These results indicate on the structure susceptible to rearrangements and fracture, compared with gels without fiber. Addition of IDFT contributed that yoghurts with both fat contents showed increased structural breakdown due to difficulty for the protein aggregates to rearrange into homogenous network after shearing as a consequence of fiber presence. Sendra et al. [11] found that particle size of fiber significantly influences on the rheological properties while Staffolo et al. [40] presented that the type of fiber is very important, in the same manner.

Textural properties of yoghurts

An important criterion for quality assessment of yoghurts is their textural properties. Textural properties of yoghurts with different fat and IDFT contents are presented in Table 4.

Our data show significant influence of fat content on the textural properties of yoghurts. It is known that milk fat positively influences the firmness, consistency and reduces whey separation of yoghurts [36]. The fat globules in homogenized milk interact positively with the protein network and acts as structure promoter [43].

On the other hand, addition of IDFT did not show significant influence on textural properties of yoghurts made with the same fat content. Fernández-García and McGregor (1997) found that addition of insoluble fibers increased the apparent viscosity of the yoghurts but without significant differences. In general, fiber addition led to gritty texture in all fiber-fortified yoghurts, except in those made with oat fiber. However, rheology and textural properties of yoghurts enriched with fibers depend on the fiber size as well as the concentration [11].

Sensory evaluation

Four-way ANOVA applied on overall quality scores showed that only the IDFT and 'fiber content'*'assessors' interaction were statistically significant ($p < 0.01$). None of the evaluated characteristics showed a statistically significant difference between samples with both fat contents ('fat content' factor) and the samples at different stages of storage. Also, 3-way ANOVA showed that the 'replication' factor was not statistically significant ($p > 0.01$) taking into account each one of the considered attributes. In the other words, milk fat content and the storage time (2 or 9 d) did not influence the sensory quality of yoghurt containing IDFT. Although the sensory profile of yoghurt was perceptibly affected by the presence of IDFT (Table 5), all of the quality scores were within the ranges of 'very good' or 'excellent' quality (i.e. > 3.5). Compared with the control samples (A0 and B0), the color of the IDFT fortified samples was described as yellowish-brown and typical for these kind of dairy products that contain whole-grain cereals. The 3% IDFT samples (A2 and B2) were visibly higher in the color intensity, as compared with A1 and B1 (1.5% IDFT). Particles of IDFT were visible on the product surface causing the occurrence of mosaic-like appearance with bright and dark color shades. This appearance was more pronounced in 3% IDFT samples. The presence of this defect significantly lowered the appearance quality scores of IDFT-fortified samples ($p < 0.01$), as compared with the control samples (Table 5). Based on the response of more than 700 young educated consumers Hoppert et al. [44] reported that affective acceptance is significantly lower for reduced-sugar yoghurt with visible fiber than for the yoghurt with inulin which is soluble and does not contribute to the color of yoghurt.

The flavor of yoghurt samples with IDFT was described by the panelists as a flavor which is pleasant and typical for whole-grain cereals dairy products (grainy-like). While the flavor of 1.5% IDFT samples was described as mild, 3% IDFT samples were perceived as slightly bitter. This negative flavor note influenced the flavor quality scores of 3% IDFT samples (the 'very good' quality range) to appear at significantly lower level ($p < 0.01$) compared with 1.5% IDFT samples (the 'excellent' quality range) (Table 5; 2-d old samples). In addition, the absence of sugar in yoghurt samples was not described as disadvantage in terms of product quality. For the yoghurts with lower sugar content Hoppert et al. [44] found that elevating flavor, which could derive from the presence of cereal particles, might be helpful to increase the general acceptance of this kind of reduced-sugar products, concluding that both cereal size and cereal taste are important factors in product formulation because of their impact on general acceptance. Due to perceived grittiness/sandiness, the texture quality was the sensory parameter most negatively affected by the presence of IDFT particles, especially in 3% IDFT samples. The texture scores of both A2 and B2 samples were significantly lower compared with the rest, but still in the range of 'very good' quality. By fortifying sweetened plain yoghurt with seven types of insoluble dietary fibers from different sources, Fernández-García and McGregor [45] found that fiber addition, primarily due to the grittiness effect, led to lower overall and texture quality scores. In an another study that included 170 participants, the texture acceptance of probiotic yoghurt fortified with 1% of passion fruit fiber was at the level of 'neither like nor dislike' at the 9-point hedonic scale [38]. These results were ascribed not only to the size of fiber particles in yoghurt, but also to the shape of fibers, which had edges like stones, causing the product to have a sandy mouth feel. Contrarily, Staffolo et al. [40] reported that more than 70% of 25 tested untrained panelists rated the texture acceptance of yoghurt fortified with 1.3% of wheat fiber with the score of 4 ('like') by using the 5-point hedonic scale. They concluded that the addition of 1.3% dietary fiber to supplement yoghurts appears to be a promising avenue for increased fiber intake, with high consumer acceptability, which is in accordance with our choice of yoghurt enriched with 1.5% IDFT.

Considering overall sensory quality scores for 2 d old samples, the 3% IDFT fortified yoghurt was assessed as the products of a 'very good' quality, while the sensory quality of the yoghurt containing 1.5% of IDFT was assessed as 'excellent'.

Antioxidant capacity of yoghurt with IDFT

In yoghurt with 2.8% milk fat and 1.5% IDFT antioxidant capacity was measured and compared with antioxidant capacity of IDFT water extract and yoghurt without IDFT by using the three most commonly used tests (Table 6). All three tests showed that addition of IDFT to yoghurt increase significantly its antioxidant activity. It increases for 8% according to TPC test and for 9.2% according to RSA and ABTS tests. Antioxidant activity of yoghurt is known in literature and depend on the ingredients used[46,] as well they add before or after inoculation[46, 47]. Different materials such as lentil[48] pomegranate peel extracts[47], wine grape pomace[49] led to increasing antioxidant activity of yoghurt. IDFT's antioxidant potential is first detail analyzed in this work which is crowned by proving increased antioxidant activity in the final food product.

Conclusions

Yoghurt enriched with innovative IDFT has been confirmed as perspective functional food. Addition of IDFT in yoghurt enhanced antioxidant activity for 8-9% and significantly influenced the syneresis level, its apparent viscosity, yield stress and thixotropic behavior, while it did not show significant influence on the textural properties of yoghurts. The overall sensory quality scores indicated that yoghurt enriched with IDFT was recognized as 'excellent', which is critical to the food market. Potential commercialization of this fiber as a novel functional component of dairy products is possible which was proven by results of its hydration capacity (higher than similar fibers), good chemical composition (rich in essential microelements) and high antioxidant activity. IDFT contribute to final food product with negligible calorific value; enhanced antioxidant activity and the total content of essential elements, with no phytates. Application of the innovative IDFT in other dairy products could offer a better insight into its optimal application. To compensate for specificities of other types of food, additional research is needed to determine if similar results would be derived within other food products.

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Table 1 Chemical analysis of triticale insoluble dietary fiber (IDFT)[1]

A)[2]	mg/g IDFT	B)[2]	%
Proteins	1.99±0.08	N	2.41±0.11
Reduction sugars	2.32±0.11	C	45.65±1.95
Starch	0.84±0.03	H	7.19±0.26
Phytates	0.00±0.0	S	0.34±0.01

C)[b]	μg/1 g fiber	C)[b]	μg/1 g fiber
Al	8.36	Li	0.02
B	3.81	Mg	3538.18
Ca	6398.17	Mn	128.68
Cd	0.05	Na	345.66
Co	0.04	Ni	0.27
Cr	0.14	Pb	0.00
Cu	7.19	Sr	3.26
Fe	134.67	Zn	76.97
K	1040.35		

[1] All experiments were carried out in triplicate. Values represent means of three replicated trials±standard deviations, Standard deviations were less than 5%

[2] A) Content of proteins, reduction sugars, starch and phytates expressed as quantity in mg per 1 g of IDFT, B) Microanalysis of IDFT, N-nitrogen, C-carbon, H-hydrogen, S-sumpor, C) ICP Analysis of IDFT.

Table 2. Characterization of triticale insoluble dietary fiber (IDFT)[1]

A)[3]			Ref. value
SC	W	5.3	
	O	2.0	
Standard characterization	WRC	4.7	
	ORC	1.6	
Specific characterization	WRC	5.3	
	ORC	2.5	
	MRC	5.5	
Acidity[2]		0.5	
pH		5.4	5.0-8.0
Moisture (%)		4.5	max 8.0
Ash (%)		2.5	max 3.0

B)[3]	Bound phenols	Free phenols	Phenols dissolved in the aqueous phase
ABTS (µmol Vitamine C/g sample)	15,856.16	57.12	1,842.47
Total phenolic content (mg/100 g IDFT)	99.0	5.0	11.3
p-cumaric acid (µg/ 1 g IDFT)	11.1	1.2	0.086
ferulic acid (µg/ 1 g IDFT)	30.1	0.44	0.15
vanillic acid (µg/ 1 g IDFT)	1.5	0.0	0.0
syring acid (µg/ 1 g IDFT)	0.0	0.0	0.0

[1] Values represent means of three replicated trials, standard deviations were less than 5% (data not shown)

[2] g of citric acid/100 mL sample

[3] A) Hydration properties of IDFT. B) Antioxidant properties of IDFT. W-water, O-oil; WRC – water retention capacity; ORC – oil retention capacity; MRC – milk retention capacity.

Table 3. Dry matter, protein contents, syneresis and rheological parameters of yoghurts with different fat and triticale insoluble dietary fiber content[1]

Yoghurt sample[2]	Dry matter (%)	Proteins (%)	Syneresis (g/100 g)	A_{up}-A_{down} (Pa/s)[3]	Yield stress (Pa)	App. Viscosity at 10/s (Pa s)
A0	10.49±0.32[a]	3.14±0.12[a]	12.44±1.30[a]	287[a]	9.54±2.45[ac]	2.90±0.14[a]
A1	12.29±0.21[b]	3.41±0.18[a]	22.04±0.75[b]	275[a]	4.79±0.67[b]	2.62±0.16[a]
A2	13.15±0.44[c]	3.61±0.20[a]	29.54±3.34[c]	134[a]	2.74±1.14[b]	1.76±0.49[b]
B0	12.01±0.08[bd]	3.28±0.12[a]	6.49±0.57[d]	595[b]	11.84±1.63[a]	5.28±0.25[c]
B1	13.23±0.17[ce]	3.42±0.11[a]	14.49±0.32[e]	456[b]	8.10±1.96[c]	4.22±0.35[d]
B2	14.35±0.57[f]	3.54±0.18[a]	16.14±0.57[e]	460[b]	7.76±1.79[c]	4.11±0.7[d]

[1] All experiments were carried out in triplicate. Values represent means of three replicated trials±standard deviations; Values with different letter within the same column are significantly different ($p < 0.05$)

[2] Abbreviations: A = 1.5 g milk fat/100 g yoghurt; B = 2.8 g milk fat/100 g yoghurt; 0 = no IDFT added; 1 = 1.5 g IDFT/100 g yoghurt; 2 = 3.0 g IDFT/100 g yoghurt.

[3] Hysteresis loop – differences in area under the upward and downward curve when plotting shear stress vs. shear rate

Table 4. Textural properties of yoghurt with different fat and triticale insoluble dietary fiber content[1]

Yoghurt sample[2]	Firmness	Consistency	Cohesiveness	Index of Viscosity
	g	g.sec	g	g.sec
A0	38.30±1.53a	995.26±45.99a	-47.19±4.13a	-120.03±11.02a
A1	36.24±4.63a	941.86±131.57a	-37.38±4.98a	-91.92±14.98a
A2	39.44±3.90a	961.27±108.36a	-35.32±3.49a	-86.19±10.70a
B0	56.02±2.76b	1483.37±70.15b	-71.07±6.47b	-176.18±15.41b
B1	62.03±5.68b	1625.23±155.22b	-75.73±8.16b	-185.05±19.30b
B2	60.19±8.14b	1595.63±227.36b	-72.91±11.77b	-178.33±26.17b

[1] All experiments were carried out in triplicate. Values represent means of three replicated trials±standard deviations, Values with different letter within the same column are significantly different ($p < 0.05$)

[2] Abbreviations: A = 1.5 g milk fat/100 g yoghurt; B = 2.8 g milk fat/100 g yoghurt; 0 = no IDFT added; 1 = 1.5 g IDFT/100 g yoghurt; 2 = 3.0 g IDFT/100 g yoghurt.

Table 5. Sensory quality scores for yoghurts fortified with triticale insoluble dietary fiber (IDFT)

Yoghurt types[1]	Overall quality[2]	Appearance	Odour	Flavour	Texture
			2 days old products		
A0	4.6 ± 0,3 ^{b}	4.8 ± 0,4 ^{b}	4.7 ± 0,4 ^{a}	4.6 ± 0,4 ^{c}	4.6 ± 0,4 ^{b}
A1	4.5 ± 0.4 ^{b}	4.6 ± 0.4 ^{a,b}	4.8 ± 0.3 ^{a}	4.6 ± 0.5 ^{b,c}	4.4 ± 0.5 ^{b}
A2	4.1 ± 0.5 ^{a}	4.3 ± 0.5 ^{a}	4.6 ± 0.5 ^{a}	4.1 ± 0.6 ^{a}	3.9 ± 0.7 ^{a}
B0	4.8 ± 0.2 ^{b}	4.7 ± 0.4 ^{b}	4.8 ± 0.3 ^{a}	4.8 ± 0.3 ^{c}	4.7 ± 0.3 ^{b}
B1	4.6 ± 0.4 ^{b}	4.6 ± 0.4 ^{a,b}	4.8 ± 0.4 ^{a}	4.6 ± 0.5 ^{c}	4.5 ± 0.4 ^{b}
B2	4.2 ± 0.5 ^{a}	4.4 ± 0.5 ^{a}	4.6 ± 0.5 ^{a}	4.2 ± 0.6 ^{a,b}	4.0 ± 0.6 ^{a}
			9 days old products		
A0	4.5 ± 0.5 ^{g}	4.8 ± 0.4 ^{g}	4.5 ± 0.7 ^{e,f}	4.4 ± 0.7 ^{f}	4.6 ± 0.6 ^{h}
A1	4.3 ± 0.5 ^{f,g}	4.4 ± 0.5 ^{e,f}	4.5 ± 0.5 ^{e,f}	4.3 ± 0.6 ^{e,f}	4.2 ± 0.7 ^{f,g}
A2	3.9 ± 0.6 ^{e}	4.3 ± 0.5 ^{e}	4.3 ± 0.7 ^{e}	3.8 ± 0.8 ^{e}	3.7 ± 0.7 ^{e}
B0	4.5 ± 0.6 ^{f,g}	4.7 ± 0.4 ^{f,g}	4.5 ± 0.6 ^{e,f}	4.2 ± 1.0 $^{e.f}$	4.7 ± 0.4 ^{h}
B1	4.5 ± 0.4 ^{g}	4.5 ± 0.5 ^{e,f,g}	4.6 ± 0.6 ^{f}	4.4 ± 0.5 ^{f}	4.5 ± 0.5 ^{g,h}
B2	4.1 ± 0.6 ^{e,f}	4.4 ± 0.5 ^{e}	4.5 ± 0.6 ^{e,f}	4.1 ± 0.7 ^{e,f}	3.9 ± 0.7 ^{e,f}

[1] Abbreviations: A = 1.5 g milk fat/100 g yoghurt; B = 2.8 g milk fat/100 g yoghurt; 0 = no IDFT added; 1 = 1.5 g IDFT/100 g yoghurt; 2 = 3.0 g IDFT/100 g yoghurt.

[2] Values are the mean ± standard deviation (N = 30 = 10 assessors x 3 replications). Values marked with the same letter within the same column and at the same level of storage are not statistically different (α = 0.01).

Table 6. Antioxidant activity of yoghurt enrichment with IDFT

Antioxidant assay	Yoghurt	Yoghurt enrichment with IDFT (1.5%)
ABTS (μmol vit C/1 g of sample)	308.44	339.6
RSA (%)	29.68	32.42
TPC (mg GAE/g)	2.68	2.92

Values represent means of three replicated trials, standard deviations were less than 5% (data not shown)

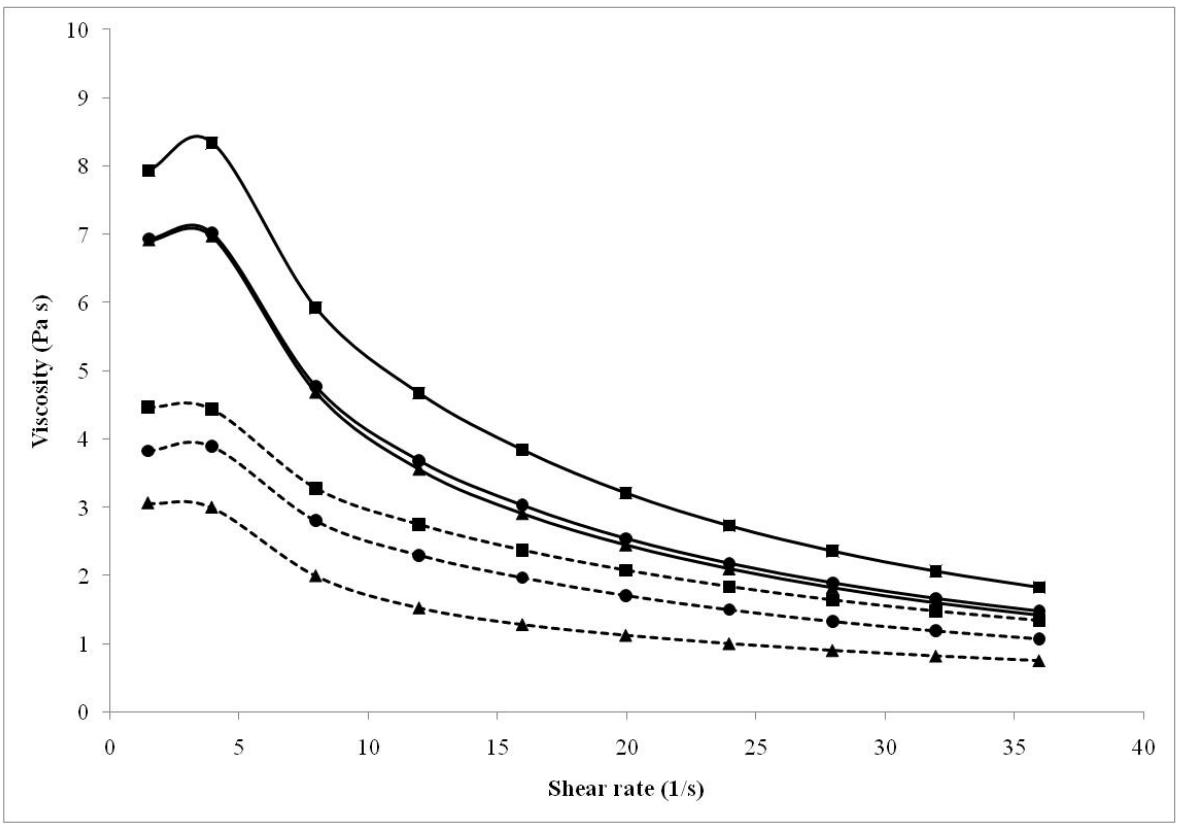

Fig. 1. Apparent viscosity of yoghurts as a function of shear rate

All experiments were carried out in triplicate. Values represent means of three replicated trials;

– Yoghurts made from milk with 1.5g milk fat/100g yoghurt fat and 0,1.5 and 3g IDFT/100g yoghurt, (■, ●, ▲, dot line, respectively) and– made from milk with 2.8g milk fat/100g yoghurt and 0,1.5 and 3g IDFT/100g yoghurt, (■, ●, ▲, solid line, respectively);

a) b)

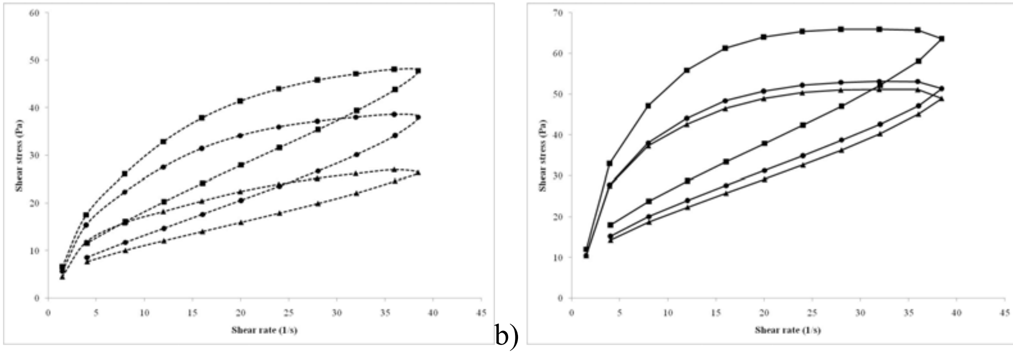

Fig. 2. Flow curves of yoghurts made with different fat and triticale fiber contents (a) yoghurts with 1.5g milk fat/100g yoghurt and 0,1.5 and 3g IDFT/100g yoghurt (■, ●, ▲, dot line, respectively) and (b) yoghurts with 2.8g milk fat/100g yoghurt and 0,1.5 and 3g IDFT/100g yoghurt (■, ●, ▲, solid line, respectively)