

## FORMULATION OF NON-FAT YOGURT WITH $\beta$ -GLUCAN FROM SPENT BREWER'S YEAST

Wided Mejri<sup>1\*</sup>, Salwa Bornaz<sup>2</sup>, Ali Sahli<sup>1</sup>

<sup>1</sup>Institut National Agronomique de Tunisie (INAT), Avenue Charles Nicolle 43, 1003 Tunis, Tunisia

<sup>2</sup>Ecole Supérieure des Industries Alimentaires de Tunis (ESIAT),  
Rue Alain Savary 58, 1003 Tunis, Tunisia

\*e-mail: wided\_mejri@hotmail.fr

### Abstract

The brewer's industry generates a huge amount of spent yeast. Its valorization can be a solution for the environmental pollution and, being rich in  $\beta$ -glucans, spent brewer's yeast are of great interest food and pharmaceutical industries.

Chemical extraction of  $\beta$ -glucan was done according to the technique described by Thammakiti *et al.* (2004) with a slight modification.  $\beta$ -glucan at appropriate concentrations (0, 0.5, 1, 1.5 and 2%), as a fat replacer was incorporated into non-fat yogurt and studied for different properties. Physico-chemical (acidification kinetic, color, physical stability) and rheological properties of the samples were instrumentally evaluated during storage at the refrigerator for 28 days. The  $\beta$ -glucan was obtained with a yield of 13.79% and a glucose purity of 96.50% (dry basis).

Results indicate that  $\beta$ -glucan had a water binding (117 g/100 g  $\beta$ -glucan), an oil binding capacity (54 g/100 g  $\beta$ -glucan), and an ability to stabilize emulsions (39% wet basis). The application of  $\beta$ -glucan in skim milk yogurt improved the rheological properties and the physical stability of the product compared to the non-fat yogurt without  $\beta$ -glucan. A proportion of 1.5% was the best proportion with a desirable attributes to that of a full fat yogurt. It had similar acidification kinetic, similar acidity (138 °D), a slightly higher syneresis (31.64% versus 30.01%), a similar water holding capacity (12.48%), a lower lightness ( $L^* = 81.93$  versus 84.41) and yellow value ( $b^* = 5.03$  versus 6.56), similar red value ( $a^* = -4.82$ ), similar values of viscosity and consistence ( $\eta = 1.30$  Pa.s,  $k = 10.48$  Pa.sn) and a lower flow index ( $n = 0.39$ ) than that of the full fat yogurt.

It is evident for this study that  $\beta$  glucan incorporation in yogurt would be an ideal fat replacer and at the same instance would benefit the consumer's with an array of health benefits. Further studies need to carry out for its application in other dairy products.

**Key words:**  $\beta$ -glucan, Spent brewer's yeast, Non-fat yogurt, Physico-chemical properties, Rheological properties.

### 1. Introduction

The brewer's industry generates a huge amount of spent yeast. With a worldwide production of beer of about 1575 million hL, 31500 - 47250 million hL of yeast accumulate each year [1]. In Tunisia, for a local production produced by "Société Frigorifique et Brasserie de Tunis" (SFBT) of 1189411 hL at the end of September 2013 [2], 23788220 – 35682330 hL of yeast accumulate. Its valorization can be a solution for the environmental pollution and also, being rich in  $\beta(1-3)$  glucans, spent brewer's yeast are of great technological interest [3].

In fact,  $\beta$ -D-glucan from *Saccharomyces cerevisiae* has been studied extensively, especially for its immunostimulatory potential and immunomodulatory activity. It could enhance innate host defenses by binding to specific macrophage receptors and activating macrophage, resulted in antitumour, antibacterial, and wound-healing activities [4], [5].

Moreover,  $\beta$ -D-glucans is poorly utilized in the human digestion tract and, therefore, function as a non-caloric food [6]. They can be used in foods as a thickener, water retention, or oil bending agent and an emulsion stabilizer [7], [8], [9], [10]. Its use in food is interesting, especially in yogurt. Being a widely spread and nutritional food, non-fat yogurt with  $\beta$ -glucan can be helpful for many people suffering from health diseases. It is well known that the amount and type of fat consumed are important to the etiology of several chronic diseases, such as obesity, cardiovascular diseases and cancer [11].

Therefore, this study has three objectives:

- Recovery of spent brewer's yeast in order to produce a value-added product -  $\beta$ -glucan.

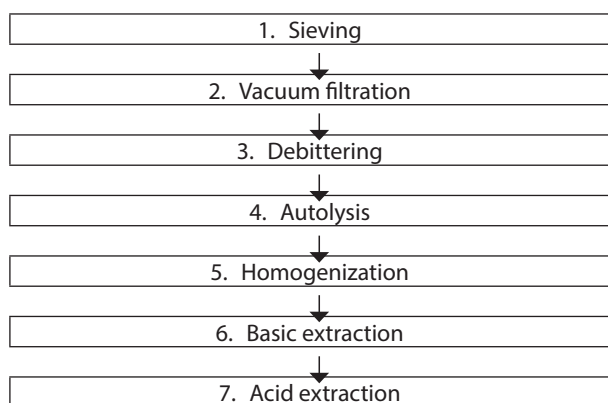
- Investigation of the effect of  $\beta$ -glucan from spent brewer's yeast as a fat replacer on the physico-chemical and rheological quality of non-fat yogurt.
- Formulation of a non-fat yogurt with  $\beta$ -glucan from spent brewer's yeast, since the fat present a major problem to human health.

## 2. Materials & Methods

### 2.1 $\beta$ -glucan extraction

The spent brewer's yeast was provided from the Tunisian brewing industry (SFBT), as a byproduct. Its solids content varied from  $20.98\% \pm 0.36$  and a pH about 5.6.

$\beta$ -glucan was extracted from spent brewer's yeast according to the chemical method described by Thammakiti *et al.* [12] with a slight modification (Figure 1). Pretreatment steps were added in order to clarify the yeast and to ensure the debittering.



**Figure 1.  $\beta$ -glucan extraction steps**

Pretreatment began by sieving yeasts through analytical sieve ( $150 \mu\text{m}$ ) and vacuum filtration. The yeast debittering using alkaline wash ( $\text{NaOH}$ ,  $\text{pH} = 10$ , at  $50^\circ\text{C}$  for 5 min.) was followed by centrifugation ( $6000 \text{ rpm}$  for 10 min. at  $4 \pm 2^\circ\text{C}$ ) and washing three times in distilled water [13]. The pH of the washed yeast was adjusted to 5 then was hydrolyzed at  $50^\circ\text{C}$  for 24 hours. A washing step is required to eliminate the chemical residue. The homogenization was carried out by cold sonication [5], [14] with an ultrasonic instrument (UC-02, Ultrasonic Cleaners JEIOTECH, Korea). Then, a basic extraction was done by adjusting pH of homogenized cell walls to 5 and adding 5 volumes of  $\text{NaOH}$  (1N) at  $90^\circ\text{C}$  during 2 hours. Finally, an acid extraction was conducted at a pH 5, with addition of 5 volumes of acetic acid (0.5N) at  $80^\circ\text{C}$  during 1 hour.

### 2.2 Physico-chemical characterization of $\beta$ -glucan

During the extraction process, the yeast and the extract were conserved at  $2^\circ\text{C}$  (AOAC 950.11) and the moisture content was determined according to AOAC 945.31C [15].

pH was measured with a pH-meter (pH 315i, WTW, Allemagne) at room temperature. Nitrogen was determined by Kjeldahl method and protein content was calculated by multiplying the rate of nitrogen by 6.25. Ash was determined by incineration in the oven at  $600^\circ\text{C}$  and fat content by Soxhlet extraction with petroleum ether [12].

The carbohydrate content is determined by subtracting from 100%, moisture, fat, protein and ash [6], [11], [16], and [17].

### 2.3 Functional properties of $\beta$ -glucan

The method used to determine the water-holding capacity, oil-binding capacity and emulsion stability were the same described by Thammakiti *et al.* [12]. The percentage of water-holding capacity was calculated as the amount of additional water held by 100 g of sample having an original moisture basis about 14%.

The Oil-binding capacity was expressed in percentage as the amount of soybean oil bound by 100 g of sample having an original moisture basis about 14% [12].

The emulsion stability was defined as the percentage of the volume of the non-separated emulsion after centrifugation, by the original volume [12].

### 2.4 Formulation of non-fat yogurt

$\beta$ -glucan at appropriate concentrations (0, 0.5, 1, 1.5 and 2%), as a fat replacer, was incorporated into non-fat (NF) yogurt and studied for different properties. The full fat (FF) yogurt was prepared as a control sample.

Yogurts were manufactured using standardized, homogenized milk provided from Professional Training Centre "Centre de formation professionnel de Cité El Khadra", as well as for the ferment and skimmed milk powder. The sugar was purchased from a local supermarket.

The  $\beta$ -glucan was dispersed into 100 mL of milk at  $60^\circ\text{C}$  with continuous stirring [18] for 10 min. then it was introduced into the total amount of milk and stirred at the same time by electric mixer for 2 seconds.

The milk was then enriched with 12% of sugar and 5% of skimmed milk powder, pasteurized at  $80^\circ\text{C}$  for 5 min. [19], cooled to  $43^\circ\text{C}$  before seeding by lactic bacteria (YO-DANISCO MIXTM LYO100DCU 496, France) and distributed over sterilized glass bottles of 100 mL each one.

The fermentation was performed in an oven temperature of about  $43 \pm 2^\circ\text{C}$  until reaching an acid value of about 90 - 95 °D [20]. Finally, yogurts were cooled and stored in a refrigerator at  $4 \pm 2^\circ\text{C}$ . Physico-chemical and rheological properties of the samples were instrumentally evaluated during storage at the day 0, 7, 14, 21 and 28 [21].

## 2.5 Physico-chemical properties of non-fat and full yogurt

The coagulation kinetics of samples was determined by monitoring the acidity every half hour, and from the fourth hour, it was done each fifteen minutes until the end of coagulation. Maximum acidification rate ( $V_m$ ) was calculated from the acidity curves according to the equation  $V_m = (d(\text{acidity})/dt)_{\text{max}}$  and expressed in absolute values ( $^{\circ}\text{D}\cdot\text{min}^{-1}$ ).  $V_m$ ; the time at which the maximum acidification rate was observed  $T_m$ , and the time at which an acidity of 90 - 95  $^{\circ}\text{D}$  was reached ( $T_e$ ) were the measured responses that characterized the fermentation kinetics [22].

Acidity was titrated using a solution of sodium hydroxide (0.1N) and phenolphthaleins [23], [24]. The pH was measured at room temperature with a pH-meter (pH 315i/ SET-A210-1012, Allemagne).

Color ( $L^*$ ,  $a^*$ ,  $b^*$ ) was measured by using a Minolta colorimeter (MINOLTA R300). In this system the  $L^*$  represent the lightness,  $a^*$  the red value and  $b^*$  the yellowness:  $+a^*$  is the red,  $-a^*$  is the green,  $+b^*$  is the yellow, and  $-b^*$  is the blue directions [21], [25].

The physical stability of the yogurt was measured by two different methods widely used in the literature: syneresis and the water retention capacity. The syneresis was determined as the amount of separated whey from 25 g of yogurt during 120 min. at 4  $^{\circ}\text{C}$  [21], [26]. Concerning of the water holding capacity (WHC), it was measured by the centrifuge method at 4500 rpm for 30 min. at 10  $^{\circ}\text{C}$  [27].

## 2.6 Rheological properties of non-fat and full yogurt

Rheological measurements were performed with a rheometer Rheomat RM180 (Standard-size DIN) with a cylindrical rotor [24]. All samples were measured at  $20 \pm 0.1$   $^{\circ}\text{C}$ , and before starting any measurements, the sample was allowed to rest for at least 2 min. [28], [29].

The studied shear rate range was between 10 and 1000  $\text{s}^{-1}$  during 120 s [30]. Rheological parameters  $K$  (consistency

coefficient) and  $n$  (flow behavior index) were determined from the power law equation (1), the most frequently used for describing the flow behavior of pseudoplastic foods [31], [32], [33], [34], [35]:

$$\sigma = K \times \gamma^n \quad \sigma = K \times \gamma^n \quad (1)$$

where  $\sigma$  is shear stress (Pa);  $\gamma$  is shear rate ( $\text{s}^{-1}$ );  $k$  is the consistency coefficient ( $\text{Pa}\cdot\text{s}^n$ );  $n$  is the flow behaviour index.

## 2.7 Statistical analysis

All the measurements were done in triplicate. Data was analyzed by ANOVA and Tukey's test ( $n = 3$ , confidence level 95%). The result was performed using the SPSS STATISTICS version 17.0.

## 3. Results and Discussion

### 3.1 $\beta$ -glucan extraction

The  $\beta$ -glucan obtained was a light-tan colored paste having a proximate composition in percentage (w/w wet basis) as followed: moisture  $80.21 \pm 0.005$ , protein  $0.10 \pm 0.01$ , ash 0.017, fat 0.12 and carbohydrate 19.55. It was obtained with a yield of 8.07% (w/w wet basis) and a glucose purity of 98.80% (dry basis).

These results cannot be compared to those of Thammakiti *et al.* [12] and Zechner-Krpan *et al.* [13] because the proximate composition expressed by these authors was determined after a step of drying, which it is not our case of study.

Compared to those of Thai team [6], [7], [8], [36], it was found that the addition of pretreatments at the method of Thammakiti *et al.* [12] induced a higher value of solids content, fat and carbohydrates in the extract and a lower value of ash and proteins (Table 1). It can be explained by the impact of the sodium hydroxide used for the debittering on the dissolution of proteins and minerals improving thus the purity (98.80% vs. 92.46% - 95.28%).

**Table 1. Comparison of physico-chemical and functional properties of  $\beta$ -glucan extracted with those of bibliography**

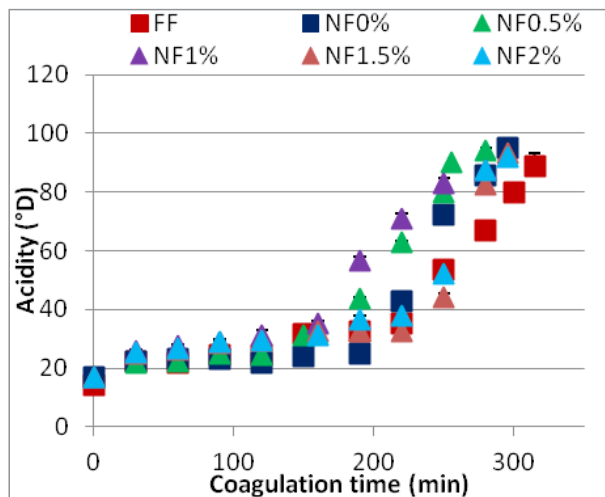
Parameters	Results	Reference 1*	Reference 2*	Reference 3*
Solid contents	$19,79 \pm 0,005$	-	6.63	6.66
Proteins	$0,10 \pm 0,01$	-	0.38	3.97
Ash	0,017	-	0.04	0.63
Fat	0,12	-	0.07	0.13
Carbohydrate	19,55	-	6.13	-
Purity*	98.80%	-	92.46%	95.28
Yield	8.07%	-	-	-
WHC	$31\% \pm 0.11$	550%	-	-
Oil binding capacity	$50\% \pm 0.19$	118%	-	-
Emulsion stability	$55\% \pm 0.30$	80%	-	-

\*Reference 1 :Thammakiti *et al.* [12] ; Reference 2 : Worrasinchai *et al.* [6] ; Santipanichwong and Suphantharika [36]; Satrapai and Suphantharika [7] ; Reference 3 : Santipanichwong and Suphantharika [8]

Concerning the functional properties of  $\beta$ -glucan, we found that it had a water holding ( $55 \pm 0.30$  g/100 g  $\beta$ -glucan), an oil binding capacity ( $50 \pm 0.19$  g/100 g  $\beta$ -glucan), and an ability to stabilize emulsions ( $31\% \pm 0.11$  wet basis) that are lower to those found by Thammakiti *et al.* [12] in the case of spray dried homogenized  $\beta$ -glucan (Table 1). This difference may be due to the initial quality of the raw material (state of development of cells) and the homogenization technique used which is different (the sonication used instead of high pressure).

### 3.2 Kinetic of coagulation

The figure 1 presents the acidification kinetic of yogurts. Two kinds of behaviors can be distinguished. The first group, consisting of NF0%, NF0.5 and NF1%, exhibited a rapid acidification. The second group, consisting of FF and NF1.5% and NF2%, exhibited a slow acidification.



**Figure 2. Coagulations' kinetics of yogurt at  $43 \text{ °C} \pm 2 \text{ °C}$  (FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%))**

The highest value of maximum acidification rate ( $V_m$ ) was observed in the case of NF yogurt with 1.5% and 2% of  $\beta$ -glucan (Table 2) after 280 min. of fermentation. These two samples had the same tendency to precipitate. This acidity increasing can be explained by the reaching of the isoelectric point (pH 4.6). The flocculated micelles precipitate forming a stable network [37], [38].

Besides, we found that in the case of a lower amount of  $\beta$ -glucan, 0%, 0.5% and 1%, the maximum acidification was reached before those with a higher percentage of  $\beta$ -glucan as for the FF sample.

Concerning the total time of coagulation, we found that the lower is the amount of  $\beta$ -glucan, the rapid is the coagulation. In comparison with the FF, we can say that the  $\beta$ -glucan at a percent of 1.5 and 2% behave as the fat do in the FF sample, encumber the matrix and

limit the circulation of lactic acid bacteria without an effect on their activities. This concord with that found by Kearney *et al.* [29] who have shown that the  $\beta$ -glucan does not affect the fermentation of lactic acid bacteria.

**Table 2. Kinetic characteristics of fermentation**

Samples	$V_m$ (°D/min)	$T_m$ (min)	$T_e$ (min.)
FF*	0.65	300	315
NF0%*	0.99	250	296
NF0.5%*	0.63	220	280
NF1%*	0.48	220	280
NF1.5%*	1.28	280	296
NF2%*	1.17	280	296

\*FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%)

### 3.3 Evolution of pH and acidity of yogurt during storage at $4 \pm 2 \text{ °C}$

After one day of storage, the acidity of yogurt samples ranged from 121 to 138°D (Table 2). These values are higher than those obtained by Kearney *et al.* [29], which are about 93°D in the case of yogurt containing  $\beta$ -glucan and about 87°D in the case of the control yogurt. They reported that these values are higher than the minimum acidity recommended by the International Dairy Federation (IDF) [29].

At a percentage of 1.5%  $\beta$ -glucan, the acidity is comparable to that of FF yogurt (Table 2). We found that these results are not in agreement with those found by Kearney *et al.* [29] who suggest that the  $\beta$ -glucan is not used by the lactic bacteria producing lactic acid and does not influence the acidity of the product.

The evolution of pH and acidity during storage at  $4 \pm 2 \text{ °C}$  was studied to test the effect of this ingredient on the viability of lactic acid bacteria in yogurt (Figure 3A). We note the tendency of the curves is similar regardless of the amount of  $\beta$ -glucan incorporated. This result is in agreement with that found by Kearney *et al.* [29].

The statistical analysis shows that only NF0% and NF0.5% yogurts had an acidity values less than that of FF yogurt after 28 days storage at  $4 \pm 2 \text{ °C}$ .

The pH gradually decreased during storage at  $4 \pm 2 \text{ °C}$ . At different proportions of  $\beta$ -glucan, the pH reached a value close to that of the control yogurt (FF) (Figure 3B).

These data are coherent with previous studies where the addition of  $\beta$ -glucans from barley or oat did not affect the growth of lactic acid bacteria in yogurt [39], and the addition of hydrocolloidal  $\beta$ -glucan from cereals in the yogurt did not significantly affect the pH during the storage [29].

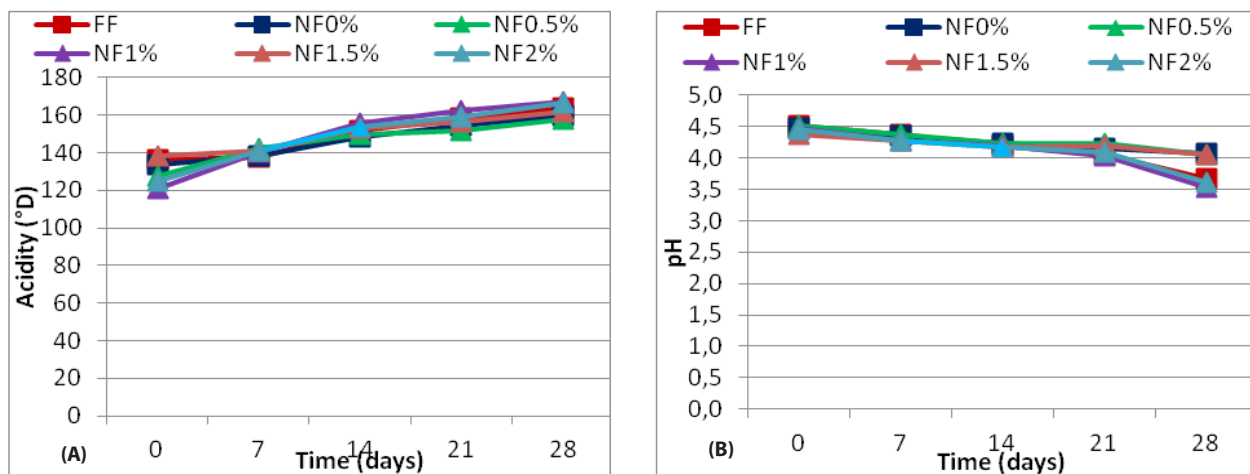
**Table 3. pH and acidity of yogurt samples after one day and 28 days of storage at  $4 \pm 2$  °C**

Samples	After 1 day of storage		After 28 days of storage	
	pH	Acidity (°D)	pH	Acidity (°D)
FF*	4.52 $\pm$ 0.006d	136 $\pm$ 1.00d	3.68 $\pm$ 0.023b	164 $\pm$ 1cd
NF0%*	4.48 $\pm$ 0.01c	130 $\pm$ 0c	4.07 $\pm$ 0.026c	160 $\pm$ 1ab
NF0.5%*	4.53 $\pm$ 0.006d	128 $\pm$ 0.58bc	4.06 $\pm$ 0.04c	158 $\pm$ 1a
NF1%*	4.48 $\pm$ 0.006bc	121 $\pm$ 1.00a	3.54 $\pm$ 0.02a	167.50 $\pm$ 0.50 <sup>e</sup>
NF1.5%*	4.38 $\pm$ 0.006a	138 $\pm$ 1.00d	4.06 $\pm$ 0.02c	161.67 $\pm$ 1.53bc
NF2%*	4.45 $\pm$ 0.025b	125 $\pm$ 3.00b	3.60 $\pm$ 0.01a	166.50 $\pm$ 0.50de

\*FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%)

Data are expressed as: Mean value  $\pm$  standard deviation

Mean values followed by the same letter in each column are not significantly different at  $P \leq 0.05$  by ANOVA and Tukey's test



**Figure 3. Evolution of acidity (A) and pH (B) during storage of yogurt samples at  $4 \pm 2$  °C (FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%))**

### 3.4 Evolution of yogurt color during storage at $4 \pm 2$ °C

The luminance of NF yogurt increased significantly with the increase of the amount of  $\beta$ -glucan from 0.5 to 1.5%.

However, it was significantly lower than that of the FF yogurt (Table 3). It is suggested to be due to the reduction of fat content [40].

The red color of NF yogurt with 1% and 1.5% of  $\beta$ -glucan was similar to that of FF yogurt. Thus, 1% and 1.5% de  $\beta$ -glucan can improve the red color of NF yogurt. While the yellowness of all NF samples remains lower than that of the FF yogurt.

No studies had been established so far about the effect of  $\beta$ -glucan on the color of yogurt. However, a sensory analysis conducted by Brennan and Tudorica [18] on the yogurt with  $\beta$ -glucan, showed that it does not affect the color at proportions of 0.5 and 1.5%.

**Table 4. Color (L\*: luminance, a\*: red value, b\*: yellowness) of non-fat yogurt with different proportions of  $\beta$ -glucan (NF0%, NF0.5%, NF1%, NF1.5% and NF2%) in comparison with the full fat yogurt (FF)**

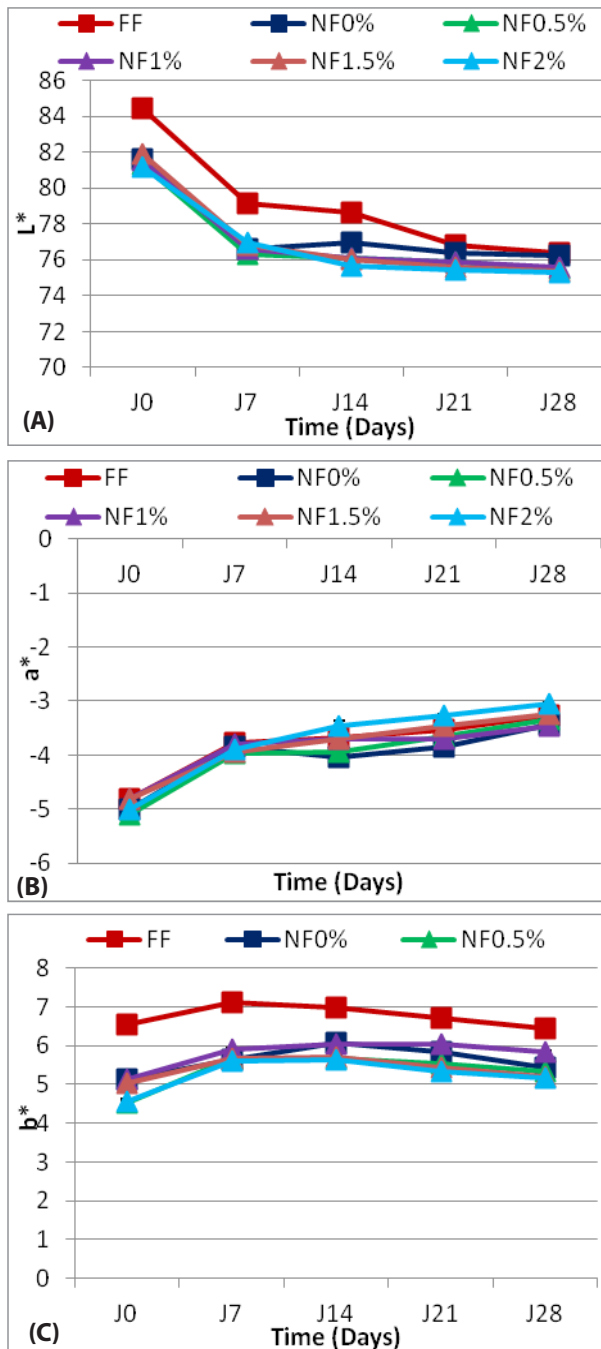
$\beta$ -glucan proportions	Color		
	L*	a*	b*
FF	84.41 <sup>e</sup> $\pm$ 0.02	-4.81c $\pm$ 0.02	6.54d $\pm$ 0.02
NF0%	81.64c $\pm$ 0.04	-5.01b $\pm$ 0.05	5.12c $\pm$ 0.05
NF0.5%	81.32b $\pm$ 0.05	-5.11a $\pm$ 0.02	4.54a $\pm$ 0.02
NF1%	81.55c $\pm$ 0.02	-4.80c $\pm$ 0.04	5.15c $\pm$ 0.03
NF1.5%	81.93d $\pm$ 0.01	-4.82c $\pm$ 0.03	5.03b $\pm$ 0.03
NF2%	81.17a $\pm$ 0.07	-5.01b $\pm$ 0.02	4.56a $\pm$ 0.01

Data are expressed as: Mean value  $\pm$  standard deviation

Mean values followed by the same letter in each column are not significantly different at  $P \leq 0.05$  by ANOVA and Tukey's test

During the storage of yogurt, we noticed that the luminance of the different samples, regardless of the amount of  $\beta$ -glucan and the fat content, decreased significantly. Throughout the storage period, the luminance of all NF yogurts with  $\beta$ -glucan was significantly lower than that of the NF0% yogurt, with the exception of the NF1.5% at the first week of storage (Figure 4).

Concerning the red color, all the samples presented the same trend. We noted that at a 1.5% of  $\beta$ -glucan, the red color was identical to that of FF yogurt at the day 0 and after 28 days of storage (Figure 4).



**Figure 4. Evolution of the color during storage at  $4 \pm 2$  °C** (FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%))

The yellowness of all samples increased at the beginning of the storage time then decreases (Figure 4). The yellowness of the NF yogurt was significantly lower than that of the FF yogurt. Only the NF1% sample presented yellowness higher than that of the NF0%.

Therefore, the storage had a significant effect on the color indices of NF yogurt.

### 3.5 Evolution of yogurt physical stability during storage at $4 \pm 2$ °C

Syneresis is the main defect of skimmed yogurt without added thickener. This parameter is of great interest to evaluate the effect of  $\beta$ -glucan. The results show that increasing the amount of  $\beta$ -glucan caused a significant decrease in the percentage of syneresis. 2% of  $\beta$ -glucan decreased the syneresis by 6% versus that of the FF yogurt (Table 5).

The addition of  $\beta$ -glucan had ameliorated the water holding capacity of NF yogurt. The physical stability of NF yogurt became similar to the FF yogurt by the incorporation of 1% of  $\beta$ -glucan. It was even ameliorated by the addition of 2% of  $\beta$ -glucan in NF yogurt (Table 5).

This result is in agreement with that found by Brennan and Tudorica [18] who found that the addition of  $\beta$ -glucan reduces syneresis of skimmed yogurt. This behavior has been attributed to the ability of concentrated barley  $\beta$ -glucan (86% of polysaccharide content) to build water in the three-dimensional network of the product. We can affirm that the  $\beta$ -glucan from spent brewer's yeast is capable of retaining water in the protein network of the product.

During storage at  $4 \pm 2$  °C, the syneresis increased while the water holding capacity was decreasing (Figure 5). Table 5 shows that all the NF samples had a higher syneresis value compared to that of the FF sample, except the NF2% yogurt which had a syneresis value close to that of FF sample. In comparison with the NF0%, from 1.5% of  $\beta$ -glucan, the syneresis was reduced.

The highest water holding capacity was obtained in the case of NF2% sample. Therefore this phenomenon justifies the intrinsic property of  $\beta$ -glucan, which is a hydrophilic compound [6].

These results correspond well with those of Brennan and Tudorica [18] and Kearney *et al.* [29]. However, Vasiljevic *et al.* (cited by Kearney *et al.* [29]) showed that the yogurt samples with 0.5% of  $\beta$ -glucan from oats or barley released substantial amounts of whey during 4 weeks of cold storage, as a result of the thermodynamic incompatibility between milk proteins and polysaccharides added. This difference in the results may be due to the diversity of  $\beta$ -glucan structures from various sources and also due to the impact of extraction processes.

**Table 5. Syneresis (%) and water holding capacity (%) of yogurt at the day 0 and day 28**

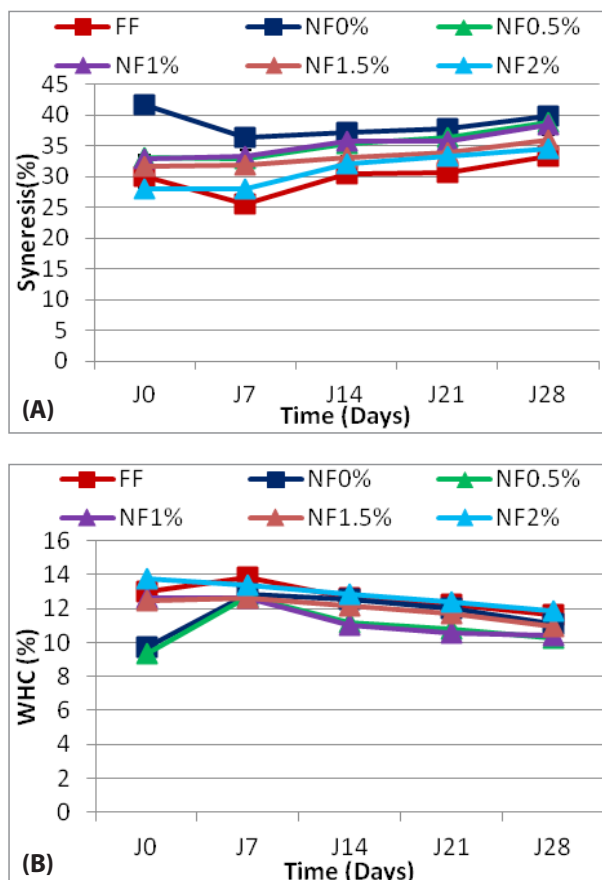
Samples	Day 0		Day 28		Day 0		Day 28	
	Syneresis	$\Delta S$	Syneresis	$\Delta S$	WHC	$\Delta WHC$	WHC	$\Delta WHC$
FF	30.01b $\pm$ 0.13	-	33.29a $\pm$ 0.90	-	13.04b $\pm$ 1.38	-	11.66cd $\pm$ 0.30	-
NF0%	41.72 <sup>e</sup> $\pm$ 0.60	39	39.75c $\pm$ 0.54	19	9.72a $\pm$ 0.25	-25	11.10bcd $\pm$ 0.53	-5%
NF0.5%	33.10d $\pm$ 0.26	10	38.82c $\pm$ 0.48	17	9.37a $\pm$ 0.18	-28	10.27a $\pm$ 0.17	-12%
NF1%	32.97cd $\pm$ 0.51	10	38.39c $\pm$ 0.74	15	12.64b $\pm$ 0.27	-3	10.45ab $\pm$ 0.28	-10%
NF1.5%	31.64c $\pm$ 0.84	5	35.92b $\pm$ 0.81	8	12.48b $\pm$ 0.24	-4	10.98abc $\pm$ 0.18	-6%
NF2%	28.10a $\pm$ 0.10	-6	34.57ab $\pm$ 0.77	4	13.78b $\pm$ 0.95	6	11.86d $\pm$ 0.15	2%

FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%)

S: syneresis; WHC: water holding capacity

Data are expressed as: Mean value  $\pm$  standard deviation

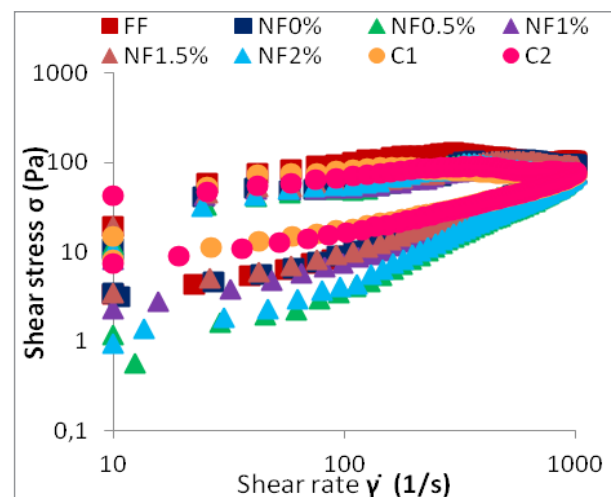
Mean values followed by the same letter in each column are not significantly different at  $P \leq 0.05$  by ANOVA and Tukey's test



**Figure 5. Evolution of syneresis (%) and WHC (%) of yogurts during storage at  $4 \pm 2$  °C**  
(FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%))

### 3.6 Evolution of yogurt rheological properties during storage at $4 \pm 2$ °C

The flow curves of yogurts prepared and also of two commercialized samples in Tunisia are presented in the Figure 6.



**Figure 6. Flow curves of yogurts**  
(FF: full fat yogurt; Nfi%: non-fat yogurt with i% of  $\beta$ -glucan (i: from 0 to 2%); C: commercialized yogurt)

From these rheograms, we find that all the samples, regardless of the percentage of  $\beta$ -glucan used, exhibit a non-Newtonian shear thinning and thixotropic behavior typical of yogurt [32], [33], [35]. Thus the addition of  $\beta$ -glucan does not affect the rheological behavior of the yogurt.

The power law model is the most appropriate to present the experimental data for different formulations, giving a minimum coefficient of determination of 0.75.

The comparison of rheological parameters of NF yogurt with those of FF yogurt showed the following facts:

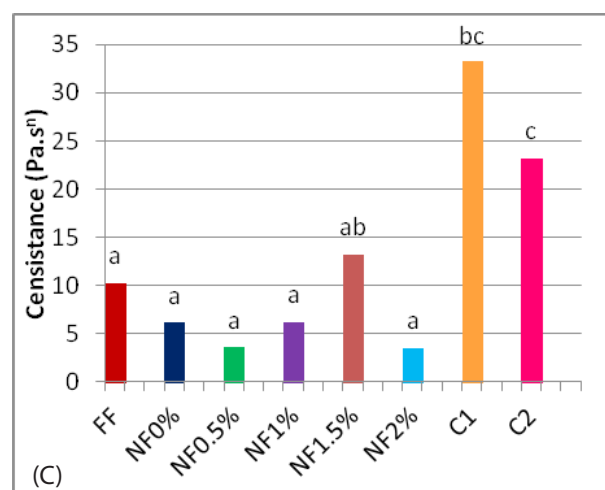
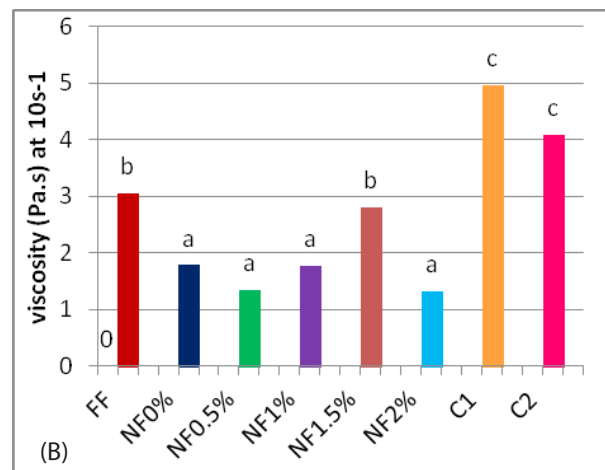
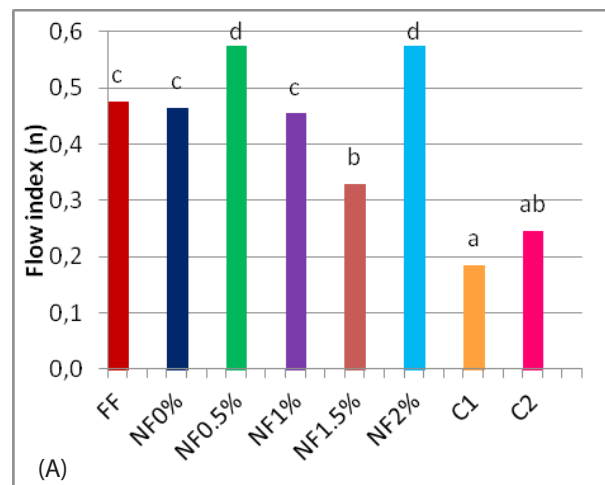
- The flow index  $n$  varied between 0.19 and 0.58 (Figure 7A), which confirms the shear thinning behavior of the yogurt. These values concord well with those found in the literature which indicated that the  $n$  values of yogurt ranges from 0.107 to 0.387 [41].
- The flow index decreased with increasing content of  $\beta$ -glucan in the yogurt. In the case of NF2 %, an inversion of the behavior was concluded (Figure 7A).
- The lowest flow index was noticed in NF1.5% yogurt and which was equal to that of the commercialized yogurt 2 (C2) (Figure 7A).
- The consistency increased with increasing the content of  $\beta$ -glucan in the yogurt. However, this increase was not considered significant except for the yogurt with 1.5%  $\beta$ -glucan (Figure 7B).
- The NF1.5% sample had a consistency close to that of the commercialized yogurt 2.
- The viscosity increased with the increase of the  $\beta$ -glucan content in the yogurt (Figure 7C). However, it is not considered significant except for the NF1.5% yogurt reaching a value similar to that of the FF sample. This result agrees well with that of Brennan and Tudorica [18].
- The evolution of the viscosity of NF yogurt due to the increase of the  $\beta$ -glucan content up to 1.5% is consistent with that found by Brennan and Tudorica [18] and Sahan *et al.* [19], who added to the NF yogurt proportions of  $\beta$ -glucan of about 0.5 ; 1 ; 1.5 ; 2 ; 2.5 and 0.25 % ; 0.5 and 1 % , respectively .
- The viscosity of all the various samples of yogurt is significantly lower than that of two commercialized yogurts (Figure 7C).
- The viscosity of the various NF samples is conform to those found by Tudorica and Brennan [18], which is between 1.2 to 4.3 Pa.s, and by Sahan *et al.* [19], which is between 2.5 and 6 according Pa.s.

Therefore, the  $\beta$ -glucan from spent brewer's yeast has a great improving of the rheological properties of skimmed yogurt at a percentage of 1.5%.

The evolution of rheological parameters during storage at  $4 \pm 2$  °C is presented in the Figure 8.

The flow index of the samples was variable over time of conservation. We noticed that it is almost stable during storage at 4 °C in the case of FF yogurt and NF1.5% yogurt. For the other NF samples, the flow index decreased during the first week of cold storage and remained virtually stable thereafter.

After 28 days of cold storage, the NF yogurts had a flow index comparable to the FF yogurt, except the NF1% sample which had a lower flow index (Figure 8 A).

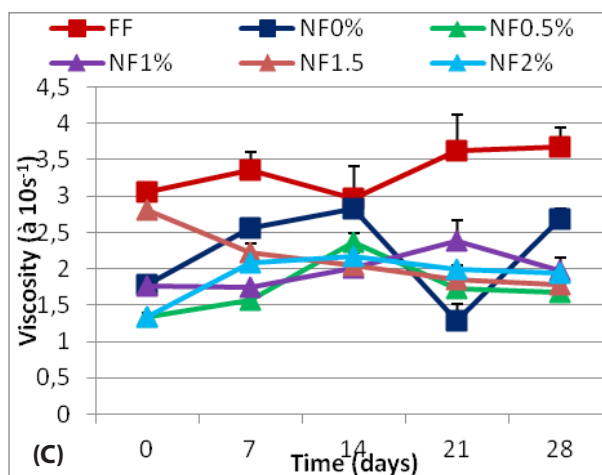
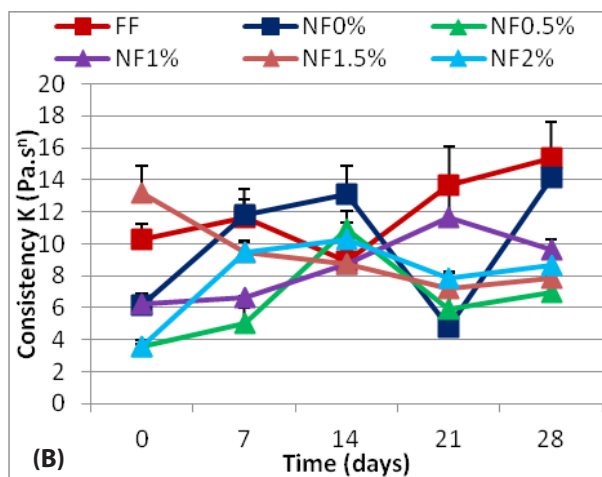
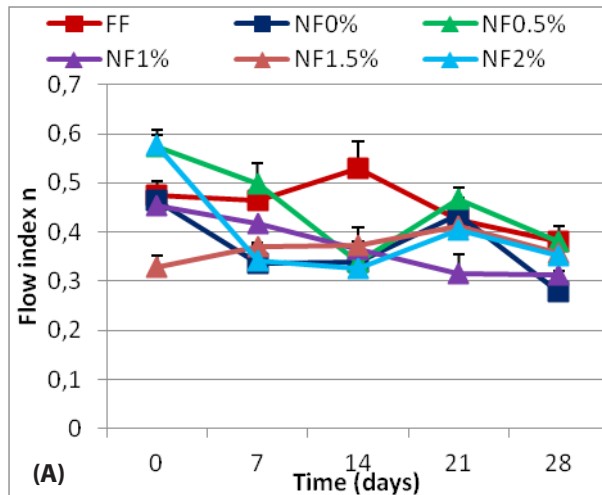


**Figure 7. Rheological properties of yogurts (flow index:  $n$ ; Consistency:  $K$ ; Viscosity:  $\gamma$ )** FF: full fat yogurt; NF*i*%: non-fat yogurt with *i*% of  $\beta$ -glucan (*i*: from 0 to 2%); C: commercialized yogurt. For each parameter, samples presented the same letter are not significantly different at  $P \leq 0.05$  by ANOVA and Tukey's test



As regards the variation of the consistency and viscosity during storage at 4 °C, three different behaviors can be distinguished (Figure 8 B). Indeed, the viscosity and consistency:

- Were stable over time during the cold storage in the case of FF yogurt.



**Figure 8. Evolution of rheological parameters of yogurt during storage at 4 ± 2 °C**  
(FF: full fat yogurt; NF*i*%; non-fat yogurt with *i* of β-glucan (*i*: from 0 to 2%))

- Increased at the beginning of storage and were stabilized from the seventh day in the case of NF1.5% yogurt.
- Increased at the beginning of storage and then decreased from the fourteenth day in the case of NF0% yogurt, NF0.5% and NF2% yogurts.

Finally, it is important to report about the rheological properties, the NF1.5% sample was the only sample with a practically stable flow, consistency and viscosity index during 28 days of storage (Figure 8).

The texture of the different samples was evaluated with a spoon, and we noticed that the samples containing 1% and 1.5% β-glucan had significantly improved the appearance of NF yogurt. This is consistent with the results found in the rheological parameters (Figure 9).



FF yogurt

NF0% yogurt



NF0.5% yogurt

NF1% yogurt



NF1.5% yogurt

NF2% yogurt

**Figure 9. Aspect of yogurt samples, evaluated with spoon after one month of storage at 4 ± 2 °C.**  
(FF: full fat yogurt; NF*i*%; non-fat yogurt with *i* of β-glucan (*i*: from 0 to 2%))

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#### 4. Conclusions

- The first purpose of this study is the valorization of spent brewer’s yeast. A slight modified chemical extraction was conducted according to the instructions of Thammakiti *et al.* [12]. Results showed that the  $\beta$ -glucan was extracted with higher purity than that found by Worrasinchai *et al.* [6], Santipanichwong and Suphantharika [36], Satrapai and Suphantharika [7] and Santipanichwong and Suphantharika [8].

- Although the obtained  $\beta$ -glucan had functional properties (water holding capacity:  $55 \pm 0.30$ g/100g  $\beta$ -glucan; oil banding capacity:  $50 \pm 0.19$  g/100 g  $\beta$ -glucan; emulsion stability:  $31\% \pm 0.11$  wet basis) lower than those of Thammakiti *et al.* [12], its incorporation into the non-fat yogurt showed motivating results.

- In the second part of this work we aimed to formulate NF yogurt with  $\beta$ -glucan. Results indicated that 1.5% of  $\beta$ -glucan could be the best proportion in order to have similar physic-chemical and rheological properties to that of the FF yogurt.

- Thus, this work highlighted the interest of the  $\beta$ -glucan use in NF yogurt. It could be further developed and completed by a sensory analysis of non-fat yogurt with  $\beta$ -glucan.

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