

## Effect of High Pressure, Autoclaving and Extrusion Treatments on Insoluble, Soluble and Total Dietary Fiber Contents of Defatted Rice Bran

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### ABSTRACT

In this study, the effects of high pressure, autoclaving and extrusion treatments on the insoluble and soluble contents and compositions of defatted rice bran were investigated. The defatted rice bran was subjected to high pressure (300, 400 and 500 MPa) for 5, 10 or 15 min, autoclaved at 122 or 136°C for 1 h and extruded with a screw speed at 100 or 140 rpm. Insoluble fiber content significantly ( $p<0.05$ ) increased when pressure increased from 300 to 400 MPa (28.62 to 32.09% at 10 min), while the increase in time affected moderately the insoluble fiber content of bran. However, autoclaved (122°C) and extruded (100 rpm) rice bran had significantly ( $p<0.05$ ) higher soluble fiber contents than others (2.87 and 3.30% respectively). The observed redistribution of soluble sugars to insoluble ones under high pressure and insoluble sugars to soluble ones occurring autoclaving and extrusion might be explained these changes on defatted rice bran fiber contents.

**Key Words:** Insoluble fiber, Soluble fiber, High pressure, Autoclaving, Extrusion

### Yağsız Pirinç Kepeğinin Çözünür, Çözünmez ve Toplam Diyet Lifi İçeriği Üzerine Yüksek Basınç, Otoklavlama ve Ekstrüzyon İşlemlerinin Etkisi

### ÖZET

Bu çalışmada yağından arındırılmış pirinç kepeğinin çözünür, çözünmez ve toplam diyet lifi içeriği ile genel bileşimi üzerine yüksek basınç, otoklavlama ve ekstrüzyon işlemlerinin etkisi araştırılmıştır. Yağsız pirinç kepeği 300, 400 ve 500 MPa basınca 5, 10 veya 15 dakika, 122°C veya 136°C sıcaklıkta otoklavlama işlemine bir saat ve 100 veya 140 d/d'lık vida hızında ekstrüzyon işlemine maruz bırakılmıştır. Basınç 300'den 400 MPa'a çıkarıldığında çözünmez lif içeriği (10 dakikalık işlemde %28.62'den %32.09'a) önemli bir oranda artmıştır ( $p<0.05$ ). Kepeğin çözünmez lif içeriğinin zamana bağlı artışı ise orta seviyede gerçekleşmiştir. Buna karşın, diğer işlemlerden elde edilenlerle kıyaslandığında, otoklavlanan (122°C'de) ve ekstrüzyona (100 d/d'da) maruz kalan pirinç kepeği daha yüksek ( $p<0.05$ ) çözünür lif içeriğine sahip olduğu bulunmuştur (sırasıyla %2.87 ve 3.30). Yüksek basınç altında çözünür şekerlerin çözünmeyenlere oranında gözlenen yeni değişim ile otoklavlama ve ekstrüzyon işlemlerindeki çözünmeyenlerin çözünenlerin oranındaki yeni dağılım, yağsız pirinç kepeği lif içeriklerindeki farklılıklarla açıklanabilir.

**Anahtar Kelimeler:** Çözünmeyen lif, Çözünür lif, Yüksek basınç, Otoklavlama, Ekstrüzyon

## INTRODUCTION

To improve the storage qualities of rice bran and to make it available all year round, it has to undergo some type of processing. During heat treatment such as autoclaving, pathogenic microorganisms are killed, antinutritional substances and enzymes inactive. However, heat treatment may have some negative effects (degradation of color, texture, etc). Today there is an increasing interests in low-thermal alternative for food preservation [1] such a high hydrostatic pressure (HHP). Many studies have shown the effects of HHP on the inactivation of microorganisms and enzymes [2], but sensory and nutritional properties have also been influenced [3]. Thus, low thermal treatment might be an attractive alternative for preservation rice bran. All these treatments influence the structure of rice bran cell-wall polysaccharides and macromolecules such as starch and protein [4]. The treatments can cause the breakage of weak bonds between the polysaccharides, as well as the cleavage of glycosidic linkages within the cell-wall polymers [5]. As a consequence of these treatments the dietary fiber content of rice bran can be affected.

Extrusion cooking is being used increasingly in the process of cereal based-foods. Cereal, as well its by-products (bran), is an important source of dietary fiber. Extrusion cooking that involves heating in combination with homogenization under high temperature and pressure during short time completely disorganizes the original structure of raw material [6]. The thermal process can make a fraction of starch less available to enzyme and the dietary fiber content [7].

Extrusion cooking treatment as well autoclaving and HHP treatments can change the structure of rice bran and vary its dietary fiber content, its composition and the ratio soluble: insoluble fractions. The purpose of this study is to investigate the effects of autoclaving, hydrostatic treatments and extrusion cooking on the soluble and insoluble fibers content of defatted rice bran and their composition on neutral sugar.

## MATERIALS and METHODS

### High Pressure Treatment

High pressure treatment was performed in a laboratory-scale, high pressure vessel (National Forge Europe, St. Niklaas, Belgium). Water was used as the pressure transmitting medium [8]. Defatted rice bran was mixed with water at ratio 1:3 and then it was submitted to HHP treatment. Treatment was performed at atmospheric pressure (0.1 MPa), 300, 400 and 500MPa for holding time of 5, 10 and 15 min at temperature 50°C. After the HHP treatment the samples were freeze-dried and stored in a desiccator until analyzed.

### Autoclaving Treatment

Defatted rice bran (200 g) was swollen in an excess of water (3 liters) for an hour at room temperature. Then, one third was directly freeze-dried. The rest was divided into two parts. One of them was autoclaved at 122°C for

1h, and the other part was autoclaved for 1 hour at 136°C. After autoclaving treatment the samples were cooled and freeze-dried [9].

### Extrusion Treatment

Extrusion was done according to the method of Kahlon et al. [10] with some modifications. Extrusion of DRB was carried out using the twin-screw laboratory extruder Thermo HAAKE PolyLab System Extruder PTW24/25D (Thermo Electron Corporation-Germany). The barrel temperature varied from feed end to die end as follow: 40-70-100°C. A feed rate of 0.7 kg/h was maintained constant while the screw speed varied 100 and 140 rpm. The initial moisture was 18% and it was equilibrated by injection water in extruder of about 0.40 kg/h. after extrusion the sample was ground and passed through sieve 0.8 mm.

### Extraction Insoluble and Soluble Dietary Fiber

Insoluble (IDF) and soluble dietary fibers (SDF) were extracted according to the method described by Mirko et al. [11]. The combined values for IDF and SDF gave a value for total dietary fiber (TDF).

### Neutral-Sugars Composition of IDF and SDF

Neutral-sugars determination of SDF was done after hydrolysis for 90 min with 1 M sulfuric acid at 100 °C [12]. Neutral-sugars in IDF were determined after prehydrolysis in 12 M H<sub>2</sub>SO<sub>4</sub> for 1h at 30°C, followed by hydrolysis of diluted solution to 1 M at 100°C for 90 min with shaking [12]. After hydrolysis the sample was centrifuged at 4000 rpm for 15 min. the residue was washed twice and the supernatant and washing water were combined for neutral-sugars determination. Reduction was done as described by Harris et al. [13] for those obtained after sulfuric acid hydrolysis of SDF and IDF. Monosaccharides were analyzed by gas-liquid chromatography (GLC). The residue was dried at 105°C overnight and gravimetrically quantified as klason-lignin. Uronic acid in hydrolysates from both SDF and IDF was quantified spectrophotometrically by the Scott [14] method using galacturonic acid as standard.

### Statistical Analysis

All the analyses were carried in triplicate, and mean ± standard deviation (SD) values have been presented. Data were statistically evaluated by one-way analysis of variance (ANOVA) using Statistical Program for Social Science (SPSS) (version 10) for significance ( $p \leq 0.05$ ) and the Tukey test at 95% confidence level.

## RESULTS and DISCUSSION

### High Pressure Treatment

The results of the effects of high hydrostatic pressure (HHP) treatments on the yields of TDF, IDF and SDF are shown on the Figure 1a, b, and c respectively and in Table 1.

### Effects of Pressure on DRB Fiber Fractions

The content of IDF and SDF was affected by the pressure to a great extent. When the pressure increased from 300 to 400 MPa for the same duration, the IDF content significantly ( $p < 0.05$ ) increased almost to 10% in 5 min (Table 1 and Figure 1b). At the same time the SDF content significantly ( $p < 0.05$ ) decreased to 10 and 39% respectively for 300 and 400 MPa (Figure 1c and Table 1). Such IDF content, TDF content also significantly ( $P < 0.05$ ) increased to 7% with an increased of pressure from 300 to 400 MPa (Table 1 and Figure 1a). Thus, the simultaneous changes in the IDF and SDF contents also resulted in the similar TDF content.

However there was no significant ( $p < 0.05$ ) change of the TDF, IDF and SDF contents when pressure increased from atmospheric pressure (0.1 MPa) to 300 MPa for 5 and 10 min (Table 1). But, for 15 min TDF and IDF contents significantly ( $p < 0.05$ ) increased, while, SDF content significantly ( $p < 0.05$ ) decreased (Table 1). When the pressure varied from 400 to 500 MPa for 5 treatment the TDF and IDF content increased significantly, but, no change was observed for 10 and 15 min treatment (Table 1). At 500 MPa, there was a tendency to an increase in the proportion of SDF at both 10 and 15 min treatment compared with 400 MPa. This might be due to a reduction of IDF.

Table 1. The dietary fiber content (g/100g dry weight) of DRB exposed to HHP treatment at different pressures during different times

Time min	Pressure MPa	Fiber Fractions*		
		TDF**	IDF	SDF
5	0.1	29.30±0.48a	27.55±0.55a	1.75±0.12b
	300	29.63±0.42a	28.05±0.40a	1.58±0.03b
	400	31.83±0.32b	30.75±0.15b	1.08±0.30a
	500	32.97±0.15c	31.97±0.11c	1.00±0.10a
10	0.1	29.32±0.55a	27.61±0.91a	1.71±0.14b
	300	30.32±0.17a	28.62±0.70a	1.40±0.18ab
	400	33.09±0.33b	32.09±0.30b	1.00±0.20a
	500	33.00±0.13b	31.99±0.70b	1.01±0.12a
15	0.1	29.51±0.18a	27.79±0.50a	1.72±0.12c
	300	30.84±0.70b	29.59±0.60b	1.24±0.40b
	400	33.13±0.11c	32.17±0.70c	0.96±0.30a
	500	33.01±0.19c	32.02±0.54c	0.99±0.60a

\*TDF: total dietary fiber, IDF: insoluble dietary fiber, SDF: soluble dietary fiber

\*\*Values in the same column for the same time followed by different letters are significantly different ( $p < 0.05$ ).

The changes in the IDF and SDF contents might be due to the change of some monomeric sugar contents. arabinose, glucose, xylose and uronic acid contents of IDF significantly increased when the applying increased from 300 to 500 MPa at the same time their contents in SDF significantly decreased (Table 4). Thus, the changes in the IDF and SDF content can be explained mainly by the redistribution of soluble uronic acids, arabinose, glucose and xylose to insoluble ones. The lower solubility of these pectic substances may be due to the inactivation of pectinmethylesterase (PME) under high pressure [15], which demethoxylates pectin and open up for the cross-linking of pectic chains [16]. Another enzyme present in the plant tissues, polygalacturonase (PG) which hydrolyses glycosidic linkages in the pectin has been reported to inactivate under high pressure [15]. Wennberg and Nyman [8] also reported that the change in IDF and SDF content during HHP treatment are due mainly to the redistribution of soluble pectins molecules such as arabinose, uronic acid and galactose to insoluble ones.

### Effects of Time on DRB Fiber Fractions

The duration of treatment can also influence the fiber fraction contents. Generally the content of SDF decreased, while that of IDF increased with increasing of applying time (Table 1). At the same pressure, the

increase of the treatment duration showed no significant ( $p < 0.05$ ) decrease of SDF content (Table 1). However, IDF content increased to 2 and 4% when the time increased from 5 to 10 and 15 min and 4% for TDF under 300 and 400 MPa. But, a tendency to decrease of TDF and IDF contents and to increase of SDF content at 500 MPa (Table 1) was observed. The redistribution of SDF to IDF also might explained the changes observed in the TDF, IDF and SDF contents of DRB. This redistribution of soluble to insoluble fiber at increasing pressure is in the contrast to previous studies where traditional processing and cooking techniques were employed [17]. It has been shown that IDF may be solubilized, degraded to smaller fragments, probably due to cleavage of glycosidic linkages and breakage of weak bonds between polysaccharides during wet heat treatment [18]. In the view of obtained data, the intensity of the pressure applying in the treatment of DRB highly affected the dietary fiber content than the applying time. High pressure results to an increase of IDF content and a decrease of SDF content and accordingly the TDF content also increased. Wennberg and Nyman [8] also concluded that the HHP treatment decreased the SDF content and increased the IDF content in the white cabbage.

**Autoclaving Treatment**

The results of the effect of autoclaving treatment on the TDF, IDF and SDF contents of the DRB are shown in the Table 2 and Figure 2. The IDF content was significantly ( $p < 0.05$ ) affected by autoclaving treatment. With increasing temperature to 122 and 136 °C the IDF decreased to 25 and 30% respectively compared to raw DRB (Table 2). At the same time the TDF contents also significantly ( $p < 0.05$ ) decreased to 23% when the temperature increased to 136 °C (Table 2).

Inversely to IDF, there was not a significant change in the TDF content, when temperature increased from 122 to 136 °C (Table 2). The SDF content significantly ( $p < 0.05$ ) increased to 87% for the autoclaved DRB (136 °C) compared to raw one. Like TDF, there was not a significant increase in the SDF content when

temperature increased from 122 to 136 °C (Table 2). These changes in IDF and SDF contents might be due the degradation of the structure of DRB. The increase of SDF might be explain by its arabinose, galactose, glucose inositol xylose and uronic acid content while these monomeric sugar contents of IDF decreased (Table 4). This change of monomeric sugar may be due to the degradation of macromolecular structures caused by autoclaving and led to the shift from IDF to SDF [19, 9]. Some of the fiber polymers also might be degraded in autoclaved product and not completely recovered during the precipitation. This simultaneous change in SDF and IDF also resulted in similar TDF. In conclusion, autoclaving treatment more significantly IDF content, but SDF and TDF content did not change significantly with increasing autoclaving temperature from 122 to 136 °C.

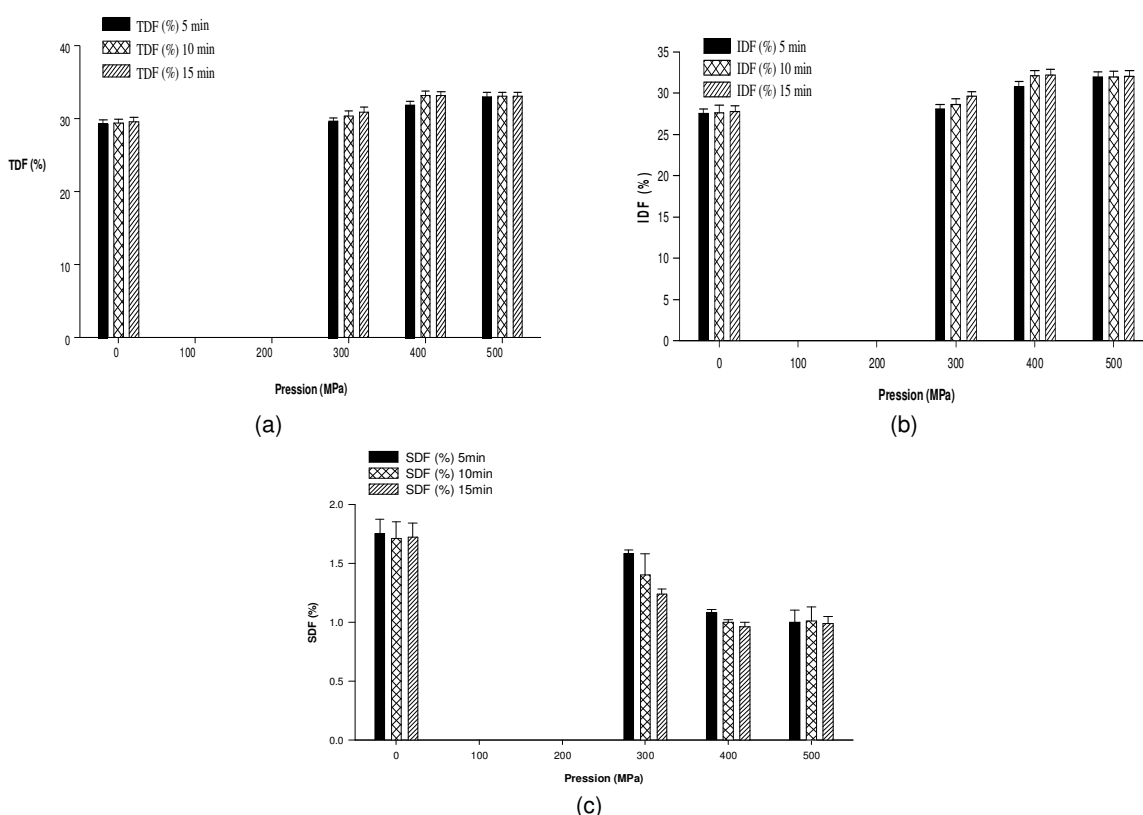


Figure1. Effects of HHP on defatted rice bran dietary fiber fractions: a, TDF; b, IDF; c, SDF

Table 2. The dietary content of defatter rice bran (DRB) exposed to autoclaving treatment

Types of DRB	Fiber Fractions (% w/w dry DRB)*		
	IDF**	SDF	TDF
Raw	27.34±0.81c	1.76±0.50a	29.10±0.20b
Freeze dried	28.06±0.40c	1.71±0.45a	29.77±0.30b
Autoclaved at 122 °C	20.54±0.29b	2.87±0.35b	23.41±0.24a
Autoclaved at 136 °C	19.14±0.27a	3.29±0.40b	22.43±0.29a

\*TDF: total dietary fiber, IDF: insoluble dietary fiber, SDF: soluble dietary fiber

\*\*The values in the same column with the same letters are not significantly different ( $p < 0.05$ )

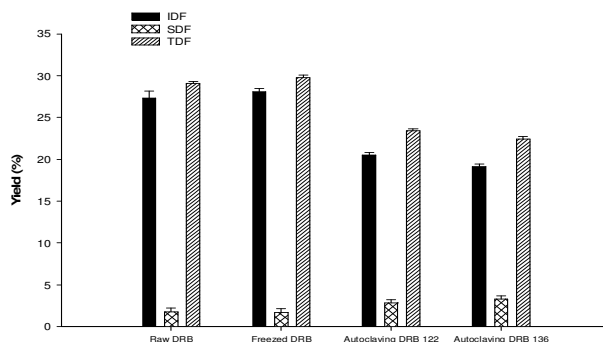


Figure 2. Effect of autoclaving on TDF, IDF and SDF fractions of defatted rice bran

### Extrusion Treatment

The effect of extrusion treatment on defatted rice bran dietary fiber (DRB) fractions is shown in the Figure 3 and Table 3. The IDF content of extruded DRB significantly ( $p < 0.05$ ) decreased to 10 and 6% for screw speed of extruder 100 and 140 rpm respectively (Table 3). Thus, extrusion processing resulted in a decrease of the IDF content compared to the raw DRB. Those values fell within the range reported in the literature for the IDF content of DRB [20]. When comparing extruded DRB samples with each another, the DRB extruded at 100 rpm and 140 rpm did not differ significantly ( $p < 0.05$ ), but both were significantly different ( $p < 0.05$ ) from the one unprocessed. This reduction was lower at high screw speed. Similar trend was observed with TDF content (Table 3). Potentially, there were several mechanisms impacting the lowering of IDF, at the same time, with one or more lowering the IDF and one or more increasing it. Some of the possible mechanisms are explained below:

- Exposure to shear stress caused strain on the IDF macromolecule resulting in the chemical bond breakage thus, creating smaller particles which were soluble [21].
- At lower screw speed, the pressure within the extruder is higher due higher screw fill. That higher pressure may have had a greater effect on the solubility of the fiber than the shear rate generated during high rpm extrusion.
- Formation of starch fractions resistant to  $\alpha$ -amylase degradation leading to increased DF due to differences in the extrusion temperatures utilized, as reported by others [22, 23].
- Complexes might have been formed between polysaccharides and lipids which could not be

cleaved by  $\alpha$ -amylase or amyloglucosidase, nor extracted by hexane and which were, thus, measured as IDF [24].

In this case, the change of IDF content of extruded DRB can be explained by these mechanisms mentioned above expected the formation the formation of complexes between polysaccharides and lipid because the used rice bran is defatted rice bran with low fat content (2.8%), thus the possibility of formation of these complexes is the lowest. SDF content of extruded DRB significantly ( $p < 0.05$ ) increased to 70 and 87% for screw speed of extruder 140 and 100 rpm respectively (Table 3). These values were higher than those reported by Prosky [20]. The difference might be due to the rice variety and the extrusion conditions. Thus, the extrusion processing increased significantly ( $p < 0.05$ ) the SDF content. The increase of SDF content can be explained by the redistribution of IDF due to several mechanisms as mentioned above [25].

TDF content of extruded DRB was decreased to 4 and 1% for 100 and 140 rpm, respectively. The TDF content of DRB decreased significantly ( $p < 0.05$ ) when extruder screw speed was 100 rpm, while it was not significantly ( $p < 0.05$ ) different for 140 rpm (Table 3). Thus, low screw speed decreased significantly the TDF content compared to high screw speed. This change in the TDF content was mainly due to the redistribution of IDF and/or mechanical decomposition of dietary fiber [26]. Thus, extrusion-cooking of defatted rice bran caused a highly decrease of IDF content and increase of SDF content while, TDF content changed moderately.

Table 3. Dietary fiber contents of DRB exposed to extrusion-cooking treatment

Treatment	Fiber fractions (g/100g dry weight DRB)*		
	IDF	SDF	TDF
Raw	27.34±0.87b	1.76±0.15a	29.10±0.77b
100rpm	24.60±0.47a	3.30±0.27b	27.90±0.36a
140rpm	25.77±0.30a	3.03±0.11b	28.80±0.18ab

\*The values in the same column with the same letter are not significantly different ( $p < 0.05$ )

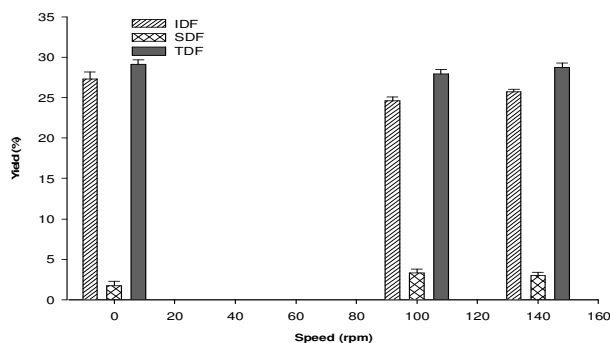


Figure 3. Effect of extrusion cooking on the yield of TDF, IDF and SDF of DRB

### Effects of different physical treatments on monomeric sugar contents of IDF and SDF fractions of DRB

The results of the effects of HHP, Extrusion-cooking and autoclaving treatments on the monomeric sugar compositions of IDF and SDF of DRB are shown in the table 4. Among the sugars of the IDF of raw DRB, the predominant sugar were glucose (25.04%) followed by uronic acid (23.90%), xylose (14.94%), Inositol (14.94%), arabinose (13.22%) and galactose (6.53%).

The sugars, included arabinose, rhamnose and uronic acid constituted up to 37% of total sugars, suggesting the presence of arabinose-rich pectic substances. The xylose-containing polymer in the IDF could possibly be a xylose rich hemicellulose comprising xylose, arabinose, galactose, glucose and mannose) [27]. The percentage of predominant sugars of SDF was uronic acid (43.18%), inositol (29.54%) glucose (10.22%), arabinose (5.68%),

galactose (4.54%), xylose (3.4%), mannose (2.27%) and rhamnose (1.13%). In the SDF the sugar such arabinose, uronic acid, galactose and rhamnose accounted for more than 50% of total sugar content, implying that SDF was mainly composed by pectic substances. On the other hand, the minor amount of monosaccharides such rhamnose, galactose, and xylose together with the significant amount of uronic acid were indicative of the presence of hemicellulose.

Autoclaving treatments of DRB have affected IDF and SDF sugar contents. In the IDF galactose and xylose contents significantly increased from 6.53% to 14.89 – 17.89% and 15.37% to 24.58 – 29.12% of total sugars respectively, while, the glucose disappeared and the mannose content was increased to 7.07% (Table 4). Other sugars did not markedly vary. In the SDF, sugar such arabinose, galactose, glucose, inositol and uronic acid significantly increased compared to SDF of raw DRB (Table 4).

Table 4. Monomeric sugar contents of IDF and SDF fractions of DRB exposed to different treatments

Monosaccharides	Fiber fractions	Monosaccharide contents (g/100g dry weight)					
		Untreated	Autoclaving		Extrusion 100 rpm	HHP	
			122°C	136°C		300 MPa	500 MPa
Arabinose	IDF	3.38	2.79	1.80	5.61	3.55	5.28
	SDF	0.10	0.20	0.35	0.25	0.077	0.037
Galactose	IDF	1.67	2.86	3.20	6.92	1.02	2.08
	SDF	0.08	0.11	0.19	0.19	0.062	0.025
Glucose	IDF	6.40	-	-	-	6.72	7.88
	SDF	0.18	0.21	0.33	0.39	0.141	0.078
Inositol	IDF	3.82	2.76	2.10	1.78	4.01	2.31
	SDF	0.52	0.96	0.72	0.69	0.384	0.375
Mannose	IDF	0.19	1.36	2.06	0.20	0.20	0.20
	SDF	0.04	0.05	0.08	0.07	0.034	0.009
Rhamnose	IDF	0.06	0.05	0.004	0.10	0.06	0.07
	SDF	0.02	0.03	0.06	0.04	0.015	0.004
Xylose	IDF	3.93	4.72	5.21	6.82	4.13	4.95
	SDF	0.06	0.08	0.18	0.11	0.043	0.018
Uronic acid	IDF	6.11	4.67	3.52	1.57	7.07	7.24
	SDF	0.76	1.23	1.38	1.56	0.65	0.45
Klason lignin	IDF	1.78	1.33	1.25	1.60	1.86	2.08
	SDF	ND	ND	ND	ND	ND	ND
<b>Total IDF/SDF</b>		27.34/1.76	20.54/2.87	19.14/3.29	24.6/3.3	28.62/1.4	32.09/1

Extruded IDF has exhibited the mainly sugar as the IDF of autoclaved DRB. However, it was found that uronic acid and inositol contents decreased, while other sugar contents increased (Table 4). SDF of extruded DRB also

exhibited the same mainly sugar as autoclaved SDF, but the glucose and uronic acid content increased significantly.

The results among the IDF from DRB treated at 300 and 500 MPa revealed that the predominant sugars were uronic acid, glucose, arabinose, xylose, inositol and galactose (more than 90% of total sugars). At 300 MPa their contents were similar to the IDF from raw bran, but they significantly increased when the pressure was 500 MPa. Arabinose, glucose, xylose and uronic acid were highly affected by the high pressure (Table 4). In the SDF, the predominant sugars were the same as SDF from raw DRB, however, the uronic acid, glucose, arabinose and galactose content significantly decreased at 500 MPa (Table 4).

Extrusion, autoclaving and HHP treatments of DRB strongly affected its IDF and SDF monomeric sugar contents. Autoclaving treatment decreased IDF arabinose, glucose, and inositol and uronic acid content while; as well extrusion, it increased SDF arabinose, galactose, glucose, inositol, xylose and uronic acid contents (Table 4). On the other hand, HHP treatment increased the IDF arabinose, galactose, glucose, xylose and uronic acid contents (Table 4).

## CONCLUSION

The study shows that HHP treatment of DRB increased its IDF content by redistribution of soluble sugars to insoluble ones, while autoclaving and extrusion increased the SDF contents. Thus, HHP treatment may be interesting with regard to the nutritional and physiological effects by promoting bulking capacity and protection against constipation and colonic cancer [28] [29]. However, the increasing of SDF contents of extruded and autoclaved bran indicated that these treatments also improve the nutritional and physiological properties such regulation of blood glucose level and blood lipid and LDL-cholesterol, thus prevent against coronary heart diseases. All these treatments can improve the use of DRB fibers as functional food.

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