

The recycling of brewer's processing by-product into ready-to-eat snacks using extrusion technology

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Abstract

Brewer's spent grain (BSG) is the main by-product of the brewing industry. The incorporation of BSG into ready-to-eat expanded products and its effects on the textural and functional properties of extrudates have been studied. Dried and milled BSG at levels of 10–30% was added to the formulation mix made of wheat flour, corn starch and other ingredients. The results obtained from the analysis of the extrudates are discussed in terms of the interaction between the ingredients and effects of processing conditions. The samples were processed in a twin-screw extruder with a combination of parameters including constant feeding rate of 25 kg/h, process temperatures 80–120 °C and screw speeds of 150–350 rpm. Pressure, torque and material temperature during extrusion were recorded. The extrudate properties of nutritional and textural characteristics were measured. Image technique investigations provided useful information on internal structure of the extruded products, total cell area, and their contribution to the appearance and texture. It was found that addition of BSG significantly increased protein content, phytic acid and bulk density, decreased sectional expansion index, individual area and total area of the cells. The higher level of BSG resulted in cells with thicker walls with a rougher surface.

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1. Introduction

The food processing industry produces large quantities of waste products. Many of these by-products are usable as raw materials for other purposes. Brewer's spent grain (BSG), the residue left after separation of the wort during the brewing process (Santos et al., 2003) is considered as a lignocellulosic material rich in around 20% protein and 70% fibre and contains about 17% cellulose, 28% non-cellulosic polysaccharides, chiefly arabinoxylans and 28% lignin (Mussatto et al., 2006). Around 3.4 million tonnes of spent grain from the brewing industry are produced in the EU every year (Eurostat data). These plant-derived waste co-products are known to contain significant amounts of valuable components, which remain unexploited waste in the current processes. Because of its high moisture and fermentable sugar content, BSG becomes an environmental

problem after a short time (7–10 days) (El-Shafey et al., 2004). It contains a high level of dietary fibre and has a strong potential for being recycled and upgraded for use within the food chain, in products such as high value food additives, composts and animal feeds.

The incorporation of BSG into food products as a source of dietary fibre has been addressed in the literature (Öztürk et al., 2002; Prentice and D'Appolonia, 1977) and it has been suggested to have a role in the prevention of certain diseases (Aman et al., 1994; Hassona, 1993; Zhang et al., 1991). This is reflected by numerous publications such as its incorporation into flour mixed breads (Finley and Hanamoto, 1980; Kawka et al., 1999; Prentice and D'Appolonia, 1977), cookies (Kissell and Prentice, 1979; Öztürk et al., 2002; Prentice et al., 1978), animal and fish feed (Batajoo and Shaver, 1994; Dung et al., 2002).

There is a growing interest to increase the dietary fibre content of extruded products by supplementing with wheat bran, corn bran, oat bran, wheat fibre, sugar beet fibre and beta-glucans (Gaosong and Vasanthan, 2000; Lue et al.,

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1991; Martianez-Tomea et al., 2004; Mendonca et al., 2000; Yanniotis et al., 2007). These types of products are potentially healthier due to the increased fibre, and can be used as a prophylactic product. In extrusion cooking, a high-fibre cereal product under mild conditions affects digestibility of starch, dietary fibre components and phytate in the stomach and small intestine of man (Sandberg et al., 1986). Thus, BSG could be a cheap source of protein and fibre that may provide a number of benefits when incorporated in human diets (Mussatto et al., 2006). Innovative ways are being developed to bring dietary fibre into new appealing high-fibre products that contribute to the recommended dietary fibre intake (National Academy of Science, 2002). The development of new techniques to use this agro-industrial by-product is of a great interest due to the large volumes of spent grain produced. However, reference searches indicate that no research on the incorporation of BSG into extruded products has been published.

The primary objective of this study was to exploit the use of food processing waste i.e. BSG and incorporate it into a ready-to-eat extruded snack. The second objective was to investigate the effects of BSG, corn starch and screw speed on selected physical and nutritional properties of the snacks. Product quality can vary depending on the extruder type, screw configuration, feed moisture, and temperature profile in the barrel, screw speed and feed rate.

2. Materials and methods

The ingredients used for the ready-to-eat snack preparation were wheat flour with composition (w/w) of 12.6% protein, 1.3% fat, 0.9% sugar, 77.3% starch and 0.9% fibre (Smiths Flours Mills, Worksop, UK); corn starch (w/w) with moisture content of 12% and composition of 80% amylopectin, 20% amylase and 88% carbohydrates (Cerestar, A Cargill Company, Manchester, UK); yogurt powder (db) with 5.5% moisture content, 4.8% protein, 1.0% fat and 88% carbohydrates (Fisue Food Ingredients, Milton Keynes, UK); onion powder (db) with 6% moisture content, 1.2% protein, 0.2% fat and 92.6% carbohydrates (Fisue Food Ingredients, Milton Keynes, UK), tomato powder (w/w) with the composition of 14.1% protein, 55.7% carbohydrates, 3% fat and 12.3% fibre (Fisue Food Ingredients, Milton Keynes, UK), yeast (Fermipan, Gist-Bracades, Holland), dill and mint (Chat Moss Herbs, Manchester, UK); paprika and salt (purchased from a local supermarket in Manchester).

A commercial sample of BSG with composition of 75% moisture, 1% ash, 4.8% protein, 16% fibre, 2.1% fat and 1.1% carbohydrates content was supplied by the Joseph Holts Brewery (Manchester, UK).

2.1. Preparation of dry BSG

The source of BSG was based on brewing of barley and hops and is the remains after extraction of the wort, before

fermentation. The BSG was refrigerated immediately after collection and then dried the following day at 150 °C for 4 h in the Teknitronic reel oven (Teknigas Ltd., Sussex, England) to a moisture content of 5–7%. The dried sample was finely milled using 1, 0.5 and 0.25 mesh screens, packed and then sealed in polyethylene bags and kept at room temperature until use. The composition of dried BSG was 20.30% protein, 53.39% fibre, 8.32% fat and 10.76% carbohydrates content.

2.2. Preparation of sample

Ingredient formulations for extrusion products are given in Table 1. Dry weight of BSG at levels of 10%, 20% and 30%, were added to each mix on a sample weight basis. All the ingredients were weighed and then mixed in a Hobart NCM mixer (Process Plant and Machinery Ltd., UK) for 20 min. Forty-six samples in total were prepared. After mixing, the samples were stored in polyethylene bags at room temperature for 24 h before extruding.

The moisture content of all the samples was estimated using the oven method (AOAC method, 1984) and later adjusted by pumping water into the extruder.

2.3. Extrusion experiments

Extrusion trials were performed using a Werner and Pfleiderer Continua 37 co-rotating twin-screw extruder (Stuttgart, Germany). The barrel diameter and *L/D* ratio were 37 mm and 27:1, respectively. A screw configuration that was a standard design for processing cereals and flour-based products was used. This screw profile was made up of conveying self-wiping elements except for a section consisting of short reverse and forwarding elements to improve mixing and apply shear to the material being extruded while restricting flow and building up pressure. The exit diameter of the circular die was 4 mm. The temperature of the extruder barrel and the moisture of the feed material were kept constant at 120 and 80 °C and 14% db, respectively. A twin-screw volumetric feeder (Rospen, Gloucestershire, UK) was used for feeding the dry mixture to the extruder and a Watson–Marlow 505 DI pump (Cornwall, UK) which was used to control the solid feed and water inputs respectively.

Table 1
Formulations for extrudate development

Formulations	Wheat flour (g/100 g)	Starch (g/100 g)	Other ingredients ^a (g/100 g)	BSG (%)
1	88	0	12	0–30
2	62	26	12	0–30
3	44	44	12	0–30
4	26	62	12	0–30
5	0	88	12	0–30

^aYogurt, onion powder, tomato powder, yeast, paprika, dill, mint and salt.

The independent processing variables included moisture content (%), feed rate (kg/h) die geometry (mm), screw speed (rpm), temperature profile °C, screw configuration. The dependant variables included die pressure (psi), material temperature (°C) and torque (%).

The process conditions were set as follows: solid feed rate of 25 kg/h, moisture content adjusted to 14% and screw speeds (150–350 rpm), two barrel temperatures were utilized as 120 and 80 °C, for two zones 80 °C at feed entry and 120 °C at die exit. Pressure, material temperature and torque were monitored during extrusion runs.

Once the extrusion response parameters, screw speed, die temperature and die pressure were constant, extrudates were cut with a sharp knife (approx. 40 cm long) as they emerged from the die and left to cool at room temperature for about 30 min, then packaged in plastic bags and stored at room temperature until analysed.

Specific mechanical energy (SME) was calculated as the total power supplied (Hu et al., 1993):

$$\text{SME (kW h/kg)} = \frac{\text{Screw speed} \times \text{Power (KW)} \times \text{Torque (\%)}}{\text{Max. screw speed} \times \text{Throughput (kg/h)} \times 100}$$

2.4. Analysis of extrudates

2.4.1. Protein content (PC)

PC was estimated from the crude nitrogen content of the sample determined by the Kjeldahl method ($N \times 6.25$), (AOAC, 1984).

2.4.2. Fat content (FC)

Total FC was determined based on a gas chromatographic technique (Gertz and Fiebig, 2000).

2.4.3. Protein in vitro digestibility (PIVD)

The samples were investigated for their in vitro protein digestibility by the modified method of Hsu et al. (1977). Fifty millilitres of aqueous protein suspension having 6.25 mg protein/ml was mixed for 60 min at 5 °C. The samples were then placed in a water bath at 37 °C and the pH was adjusted to 8.00 using 0.1 N NaOH and/or 0.1 N HCl, while stirring. Lyophilized, crystallized trypsin (Sigma Chemical Co., St. Louis, MA, USA) at a concentration of 1.6 mg/ml was maintained in an ice bath and the pH was adjusted to 8.00 with 0.1 N NaOH and/or 0.1 N HCl. Five ml of enzyme solution were then added to the protein suspension, which was being stirred at 37 °C. The trypsin had an activity of 13,766 BAEE units/mg protein. A rapid decline in pH was observed. The pH drop was recorded 15 s after enzyme addition and at 1-min intervals for 10 min. Triplicate analyses were performed for each sample. The enzyme solution was freshly prepared before each series of the test. The percentage of protein

digestibility (Y) was calculated by the equation:

$$Y = 210.4 - 8.1x,$$

where x is the change in pH after 10 min.

2.4.4. Phytic acid (PA)

PA was measured by a colorimetric method according to Haugh and Lantzsch (1983). PA in the sample was extracted with a solution of HCl (0.2 N) and precipitated with a solution of Fe III (ammonium iron (III) sulphate · 12H₂O).

2.4.5. Total antioxidant capacity (TAC)

TAC of samples was determined according to the method given by Erel (2004). This method is based on the decolourization of 2,2-azino-bis (3-methylbenzothiazoline-6-sulfonate) (ABTS) radical action. The samples were extracted using 50% methanol.

2.4.6. Total phenolic compounds (TPC)

TPC was determined according a modified method of Skerget et al. (2005). This was used to calculate the concentration of total phenolics in the samples as mM gallic acid/g dry sample using a UV spectrometer, based on a colorimetric oxidation/reduction reaction. The oxidizing agent used was Folin–Ciocalteu reagent. The samples were extracted with pure methanol at 40 °C.

2.4.7. Fibre content

The total dietary fibre content of extrudates was determined using a combination of enzymatic and gravimetric methods (Sigma-Aldrich Inc., St. Louis, MO, USA) (AOAC, 1997). Milled and dried samples were gelatinized with heat stable α -amylase and then enzymatically digested with protease and amyloglucosidase to remove protein and starch present in the sample. Ethanol was added to precipitate the soluble dietary fibre. The residue was then filtered and washed with ethanol and acetone. After drying, the residue was weighed. Half of the samples were analysed for protein and the others ashed. Total dietary fibre is the weight of the residue less the weight of the protein and ash.

2.4.8. Colour

The colour of the extrudate was assessed using a Hunter-Lab colorimeter (Optical Sensor, Hunter Associates Laboratory Inc., Reston, VA, USA). In the Hunter-Lab colorimeter, the colour of a sample is denoted by the three dimensions, L^* , a^* and b^* . The L^* , a^* and b^* readings were then taken from a digital display unit (Model D 25-2). The L^* value gives a measure of the lightness of the product colour from 100 for perfect white to zero for black, as the eye would evaluate it. The redness/greenness and yellowness/blueness are denoted by the a^* and b^* values, respectively. The colour of the samples was measured after grinding the samples to fine flour (particle size <200 μm) using a type ZM100 ultra centrifugal mill (Retsch GmbH Co., Haan, Germany).

2.4.9. Water absorption index (WAI) and water solubility index (WSI)

WAI and WSI were determined according to the method developed for cereals (Anderson et al., 1969). The ground extrudate was suspended in water at room temperature for 30 min, gently stirred during this period, and then centrifuged at 3000g for 15 min. The supernatant was decanted into an evaporating dish of known weight. The WSI was the weight of dry solids in the supernatant expressed as a percentage of the original weight of sample. The WAI was the weight of gel obtained after removal of the supernatant per unit weight of original dry solids.

2.4.10. Bulk density (BD)

BD (g/cm^3) was calculated according to the method of Alvarez-Martinez et al. (1988):

$$\text{BD} = \frac{4m}{\pi d^2 L}$$

where m is the mass (g) of a length L (cm) of extrudate with diameter d (cm).

2.4.11. Sectional expansion index (SEI)

SEI was determined according to Alvarez-Martinez et al. (1988) and is defined as

$$\text{SEI} = \left[\frac{D_e}{D_d} \right]^2,$$

where D_e is the diameter of the extrudate and D_d the diameter of the die.

2.4.12. Texture analysis

Mechanical properties of the extrudates were determined by a three-point bend method using a TA-XT2 texture analyser (Stable Micro Systems Ltd., Godalming, UK) equipped with a 25 kg load cell. An extrudate 100 mm long was compressed with a probe 90 mm long at a crosshead speed 3 mm/s to 4 mm of original diameter of the extrudate. The compression generated a curve with the force over distance. The highest value of force was taken as a measurement for hardness.

2.4.13. Image analysis

Image analysis was conducted on the samples containing 50% starch and 50% wheat flour and BSG levels between 0% and 30% for each screw speed. Cylindrical shape extrudates were cut using a blade to obtain a product 10 mm in length. Images of cross sections of extrudates were captured with a $5 \times$ magnification using a digital camera, Sony, MVC-CD500, (Sony Electronics Inc., NJ, USA). Image analyses were performed using the UTHSC-SA Image tool program (Version 2.0, University of Texas Health Science Centre, and San Antonio, Texas, available by anonymous FTP from maxrad6.uthscsa.edu). Analyses were carried out on sub images of 500×500 pixels representing 422 mm^2 of the slice area and processed into grey level (8 bits). The number and cross-sectional

area of the cells in extrudates were extracted. Cell density was calculated by dividing cell numbers by cross-sectional area.

2.5. Reproducibility

All the experiments were replicated, so that the data in the paper are all mean values of three tests. The coefficient of variation (CV) of all the tests was lower than 10%.

2.6. Statistical analysis

Statistical analyses were conducted using a commercial statistical package, Design-Expert version 6.01 (Statease Inc., Minneapolis, USA). Statistical significant difference between values was evaluated with a multiple comparison test using SPSS 12.0.1 (SPSS Inc., Chicago, Illinois, US).

3. Results and discussion

3.1. Process conditions

All the data obtained during extrusion runs and SME are presented in Table 2. SME is a good quantitative descriptor in extrusion processes, since it allows the direct comparison of different combinations of extrusion conditions such as screw speed, feeding rate and torque (Moraru and Kokini, 2003). In this study, a constant solid feed rate, water rate and barrel temperature have been used. Results clearly showed that SME was highly correlated to screw speed ($r = 0.9$) and slightly related to the torque ($r = -0.6$) and pressure ($r = -0.6$). It has been found that as screw speed increases from 50–150 rpm the SME for barley extrudates linearly increased (Baik et al., 2004). It is suggested that an increase in SME was due to the increase in shear rate when screw speed was raised.

3.2. Effect of starch and BSG content on crude protein, fat and fibre levels

Analyses of the finished products (Table 3) indicated a wide range of crude PCs between 5.1% and 15.5%. Significant differences ($P < 0.005$) were noted: samples without BSG generally displayed the lowest PC across 19 different formulations (5.1–11.3%) while samples with the highest level of BSG displayed the higher level (7.1–15.5%). The PC mainly came from BSG and wheat flour, with a lowering of the PC as wheat flour was replaced with starch.

Variation in FC of the extrudates was between 0.2% and 1.9%, with a significant difference between different formulations ($P < 0.0001$) (Table 3). The FC was highly correlated to level of BSG ($r = 0.9$) but not to the level of starch. It is known that fat interferes significantly with starch gelatinization at a constant screw speed (Lin et al.,

Table 2

Process conditions recorded during each experimental run: experimental runs were obtained using Design Expert program

Starch (%)	BSG (%)	Screw speed (rpm)	Torque (%)	Material temperature (°C)	Pressure (bar/psi)	SME (kW h/kg)		
0	0	150	64.0	130.0	50.0	0.07		
		250	52.0	130.0	20.0	0.10		
		350	43.0	130.0	0.0	0.11		
	10	150	150	82.0	145.0	57.5	0.09	
			250	60.0	152.5	22.5	0.11	
			350	50.0	150.0	3.0	0.13	
		20	150	150	62.0	135.0	45.0	0.07
				250	51.0	142.5	30.0	0.10
				350	40.5	140.0	7.5	0.11
30	150	150	70.0	140.0	52.5	0.08		
		250	58.0	150.0	30.0	0.11		
		350	48.0	155.0	15.0	0.13		
30	0	200	69.0	140.0	15.0	0.10		
		300	50.0	142.5	0.0	0.11		
		300	63.0	150.0	45.0	0.10		
	10	200	200	54.0	155.0	12.5	0.12	
			300	58.0	147.5	35.0	0.09	
			300	50.0	152.5	15.0	0.11	
	20	200	200	57.0	145.0	30.0	0.09	
			300	50.0	150.0	15.0	0.11	
			300	50.0	150.0	15.0	0.11	
	50	0	150	82.0	142.5	27.5	0.09	
			250	64.0	147.5	10.0	0.12	
			350	49.0	145.0	0.0	0.13	
10			150	150	71.0	145.0	42.5	0.08
				250	61.0	145.0	5.0	0.12
				350	50.0	145.0	0.0	0.13
20		150	150	67.0	140.0	40.0	0.08	
			250	56.0	147.5	20.0	0.11	
			350	46.0	150.0	0.0	0.12	
		30	150	150	63.0	137.5	42.0	0.07
				250	49.0	142.5	15.0	0.09
				350	43.0	145.0	0.0	0.11
70		0	200	64.0	145.0	5.0	0.10	
			300	64.0	145.0	10.0	0.10	
			300	50.0	145.0	0.0	0.11	
		10	200	200	58.0	140.0	0.0	0.09
				300	50.0	142.5	0.0	0.11
				300	69.0	150.0	40.0	0.10
	20	200	200	50.0	175.2	12.5	0.11	
			300	50.0	175.2	12.5	0.11	
			300	50.0	175.2	12.5	0.11	
100	20	150	72.0	150.0	140.0	0.08		
		250	58.0	142.5	0.0	0.11		
		350	73.0	142.5	15.0	0.08		
	30	150	150	73.0	142.5	15.0	0.08	
			250	58.0	145.0	2.5	0.11	
			350	50.0	145.0	0.0	0.13	

1997); with a more pronounced decrease at higher screw speeds. One explanation is that the lubricating effect of fat during extrusion reduces the frictional torque and, consequently, the mechanical energy input (Moraru and Kokini, 2003).

As a result of adding up to 30% BSG, a wide variation in fibre content of the extrudates was observed, with values ranging between 1.1 and 14.4 (Table 3). The per cent of fibre was highly correlated to BSG addition ($r = 0.9$) with $P < 0.0001$. Other researchers have increased fibre levels in extrudates using different sources such as oat

(Martianez-Tomea et al., 2004), wheat (Martianez-Tomea et al., 2004; Wang et al., 1993; Yanniotis et al., 2007) and barley (Baik et al., 2004; Berglund et al., 1994) but no published work has been identified for BSG addition to extruded products. However, BSG has been used as a fibre source to increase dietary fibre content of bread (Finley and Hanamoto, 1980; Kawka et al., 1999; Prentice and D'Appolonia, 1977), and cookies (Kissell and Prentice, 1979; Öztürk et al., 2002; Prentice et al., 1978). In our study, compared to the effect of increasing the BSG into the formulation, the addition of starch had very little effect on fibre levels.

Table 3
Nutritional analyses of extrudates obtained from 19 formulations

Starch (g/100 g)	BSG (g/100 g)	Protein (g/100 g)	Fat (g/100 g)	Fibre (g/100 g)	Phytic acid (mg/100 g)
0	0	11.3 ^{a,g,l,e,b,c}	0.5 ^{a,b,c,i}	1.1 ^a	1004 ^a
	10	12.1 ^b	0.7 ^{b,f,g,c,l,o}	6.1 ^{b,r,e,f,n,i,k,l,n}	1176 ^b
	20	12.2 ^c	1.3 ^{d,f,h,j,n,c}	11.3 ^{c,d,e,f,g,i,f,k,l,m,n,o}	1245 ^d
	30	15.5 ^d	1.9 ^{e,k}	13.4 ^{d,e,e,f,g,j,l,m,o}	1610 ^c
30	0	10.3 ^c	0.4 ^{a,b,i,l}	2.8 ^{a,p,b}	980 ^a
	10	13.1 ^{f,b,c}	1 ^{f,g,i,l,m,n,o,t}	10.1 ^{e,b,c,d,f,g,h,i,k,l,m,n,o}	1143 ^b
	20	10.7 ^{g,k,l,p,e}	0.9 ^{g,b,l,j,l,m,n,o,c}	9.7 ^{f,b,c,d,e,g,h,i,k,l,n}	1276 ^d
	30	13.4 ^{h,c,f}	1.4 ^{h,k,n}	13.9 ^{g,c,d,e,f,j,l,m,o}	1543 ^c
50	0	8 ⁱ	0.3 ^a	2.0 ^a	1003 ^a
	10	7.9 ^{g,m,n,i}	0.7 ^{i,l}	7.3 ^{h,b,e,f,i,k,e,n}	1100 ^b
	20	9.6 ^{k,e}	1.1 ^{j,l,n,o,c}	9.1 ^{i,b,c,e,f,h,k,l,n}	1276 ^d
	30	10.8 ^{l,e}	1.6 ^k	14.3 ^{i,c,d,g,m}	1587 ^c
70	0	6.6 ^m	0.2 ^a	5.2 ^a	996 ^a
	10	8.1 ^{n,o,i}	0.7 ^{l,m,o}	7.3 ^{k,p,b,c,e,f,h,i,l,n}	1090 ^b
	20	7.5 ^{o,m,i}	1 ^{m,n,o,c}	9.7 ^{l,b,c,d,l,f,g,h,i,k,m,n}	1250 ^d
	30	9.6 ^{p,k,l}	1.3 ^{n,c}	14.4 ^{m,c,d,e,g,j,l,o}	1545 ^c
100	0	5.1 ^q	0.5 ^{a,b,i,l}	2.5 ^{a,r}	986 ^a
	20	6.7 ^{r,m}	0.8 ^o	9.0 ^{n,b,c,l,f,h,i,k,l}	1245 ^d
	30	7.1 ^{s,m,n,i}	1.2 ^c	14.0 ^{o,c,d,e,f,j,m}	1600 ^c

Values bearing the same letters within the same column are not significantly different from each other ($P < 0.05$).

3.3. Effect of starch and BSG content on TAC, TPC, PA and PIVD

The results obtained from PIVD, TAC, TPC and PA measurements are presented in Table 3. It was expected that BSG addition would effect these parameters as has been reported in several studies for different grains: durum wheat bran as an antioxidant source (Onyeneho and Hettiarachchy, 1992); sorghum (Beta et al., 2001), wheat germ/bran level (Bilgili et al., 2006), oat (Collins, 1989), oat and wheat (Martianez-Tomea et al., 2004) as sources of TPC with antioxidant activity; wheat bran fibre (Bilgili et al., 2007) as a source of PA; and corn, maize, millet and sorghum (Dahlin and Lorenz, 1993, Duodu et al., 2002) for improving in vitro protein digestibility.

Table 3 shows a significant increase ($P < 0.0001$) in PA of the extrudates as the level of BSG was increased ($r = 0.9$). In a study of fermented products (Bilgili et al., 2006) it has been shown that the level of PA is reduced as a result of its degradation, which is thought to be due its utilization by yeast during fermentation (Lasztity and Lasztity, 1990). Extrusion cooking did not change the content of PA and dietary fibre in extruded bran products compared with non-extruded products (Sandberg et al., 1986), indicating clearly that PA is affected by the process that is applied.

The use of BSG as a source of phenolic acids has been reported in the literature (Athanasios et al., 2006; Bartolomé et al., 2002; Mussatto et al., 2006). Bartolomé et al. (2002) after basic hydrolysis of the BSG, found major phenolic compounds such as ferulic and *p*-coumaric acids. In our study no difference was detected between all the

samples in terms of TPC (1.8–2.2 mM Trolox equiv/g) and TAC (9.7–12 mM Trolox equiv/g) as the level of BSG was increased. There is strong evidence that the predominant source of antioxidant activity derives from phenolic compounds (Beta et al., 2001).

Protein digestibility is an important factor when assessing the protein quality and nutritional status of a food product (Hsu et al., 1977). It was found that the sample without BSG gave lower values during incubation probably as a result of lower PC. The digestibility of proteins is the primary determinant of the availability of amino acids which is important in evaluating its nutritive quality (Paquet et al., 1987). The extrusion process has the potential to change digestibility through a combination of shearing, heat and pressure (Wen et al., 1990). It has been shown that gelatinized maize starch slightly reduced protein digestibility (Duodu et al., 2002) and the beneficial effect of different extrusion process conditions for different cereals resulted in improved protein digestibility (Bjork et al., 1983; Coulter and Lorenz, 1991; Dahlin and Lorenz, 1993). However, our study showed that the % of PIVD (69.1–76.6 g/100 g) in different formulations was not related to the level of BSG and extrusion conditions.

Different starch level in our study had no effect on TAC, TPC, PA and PIVD.

3.4. Effect of starch, BSG content and screw speed on WAI and WSI

WAI and WSI were used to estimate the functional characteristics of the products. Results of WAI and WSI

are presented in Table 4. In this study, only WAI was related to the level of BSG ($r = -0.54$) with a significant difference of $P < 0.005$, but was not related to different levels of starch and screw speed. This is not consistent with the findings of Baik et al. (2004) in the production of expanded cereals using barley flour, where increased screw speed resulted in increased WAI in extrudates. These authors concluded that increased SME by the elevated screw speed probably promoted the molecular disruption of barley flour components, leading to the increase in WAI along with the increased expansion of the extrudates. It has been reported that a higher value of WAI in the gelatinized sample is due to the presence of undamaged long polymer chains (Gomez and Aguilera, 1984). WAI decreases as the length of the polymeric chains is reduced.

In this study WSI was not related to the different level of starch content, BSG and screw speed.

3.5. Effect of starch, BSG content and screw speed on colour (L^* , a^* and b^*)

The colour values recorded for the extrudates containing different levels of starch and BSG at three screw speeds are summarized in Table 4. Results obtained with the sample containing 50% starch with different levels of BSG and screw speeds showed the significant influence ($P < 0.05$) of BSG L^* values in all samples. The samples without BSG displayed significantly higher L^* values than other samples.

Increasing the level of BSG resulted in lowering the brightness. L^* was significantly correlated ($P < 0.05$) to different levels of BSG ($r = -0.85$).

The redness (a^*) of extrudates indicated that samples containing 20–30% BSG were more negative, and therefore possibly more desirable. Redness of the product was related to the level of BSG ($r = 0.7$) with significant difference of $P < 0.05$.

Samples containing higher levels of BSG showed lower value of b^* . A negative correlation was detected between b^* and BSG ($r = -0.9$) at significant difference $P < 0.0001$.

From the data obtained it appears that different levels of starch and different screw speeds had no effect on the colour of extrudates.

3.6. Effect of starch, BSG content and screw speed on hardness, BD and expansion

The expansion, hardness and BD of the product at different formulations are given in Table 4.

The maximum force obtained has been taken as a measure of hardness of the products (Table 4). It has been shown that fibre increases the hardness of the extruded products (Mendonca et al., 2000; Yanniotis et al., 2007) as a result of its effect on cell wall thickness. In many cases, a high hardness value is not a desirable attribute for

expanded snacks. On addition of corn starch hardness of the extrudates, in this work, was reduced with an increase in softness occurring as corn starch levels increased. There was a significant difference in hardness detected between different samples with a correlation to the level of corn starch ($r = -0.6$).

The results obtained for BD of the products (Table 4) showed a weak correlation as the level of corn starch increased ($r = -0.4$) and BSG increased ($r = 0.45$).

The sample containing wheat flour and different levels of BSG (0–30%) had reduced expansion at BSG levels of 20% and 30% but not at 10%. Some authors (Faubion and Hosney, 1982; Linko et al., 1981) have reported a significant influence of the amount and type of protein present on expansion and textural properties of wheat flour extrudates resulting, with a decrease in expansion observed as gluten PC increases. Increasing the level of BSG in bread led to a decrease in loaf volume, increased water absorption and protein level (Finley and Hanamoto, 1980; Prentice and D'Appolonia, 1977). In cookie formulation it was found that maintaining acceptable physical qualities of the product with 40% BSG was possible (Kissell and Prentice, 1979; Öztürk et al., 2002; Prentice et al., 1978). At 30% and 50% starch replacement, a similar trend was detected resulting in a decrease in expansion as screw speed and level of BSG increased. This is consistent with the findings of Chiang and Johnson (1977) and Owusu-Ansah et al. (1983) that a higher screw speed resulted in a lower expansion ratio. This is thought to be due to a lower level of starch gelatinization or molecular degradation of amylose and amylopectin as screw speed affected residence time. The higher fibre content probably contributed to the lower expansion of extrudates while increasing the level of starch resulted in increased expansion. The expansion was not correlated to the addition of BSG or different screw speed. The sample containing 10% BSG did not go through extrusion process at a screw speed of 250 rpm. A similar problem was found with samples containing 10% and 20% BSG at a screw speed of 350 rpm. It has been shown (Yanniotis et al., 2007) that wheat fibre addition to starchy products decreased radial expansion in extrudates similar to the results obtained in this study. The formulation was a complex system containing different levels of starch, wheat flour and BSG, with a range of components at different levels e.g. fibre, fat, PC. In a study by Grenus et al. (1993), both radial and axial expansion of extrudates increased with the addition of 10% rice bran and decreased at higher levels. Shear strength increased with bran levels over 10% and with increase in screw speed: an examination of the puff microstructure showed an increase in structural organization with 10% rice bran and decreased cell size and incomplete cells at higher levels of bran addition.

Generally different screw speeds did not affect the expansion of the product and the BD, but were slightly related to the hardness of the products ($r = -0.4$).

Table 4
Mean values of functional and textural characteristics of extrudates

Starch (%)	BSG (%)	Screw speed (rpm)	WAI (ggel/g)	WSI (%)	Colour			Hardness (N)	SEI	Bulk density (g/cm ³)
					L*	a*	b*			
0	0	150	7.7	4.6	67.3	-3.8	31.8	20.4	7.3	0.4
	0	250	7.4	7.0	67.2	-2.8	31.3	18.5	9.3	0.2
	0	350	7.1	9.1	66.9	-3.3	30.9	19.4	8.3	0.2
	10	150	5.7	9.7	57.5	-1.1	24.2	17.0	7.8	0.3
	10	250	5.6	12.1	55.7	-1.5	23.3	15.5	8.8	0.2
	10	350	5.4	15.7	56.8	-1.7	23.4	12.4	8.5	0.1
	20	150	6.3	6.3	51.6	-1.5	20.3	16.7	4.0	0.5
	20	250	6.0	9.0	50.8	-1.7	19.6	17.2	4.5	0.4
	20	350	6.0	10.9	50.9	-1.3	20.2	13.8	4.5	0.3
	30	150	4.9	6.1	52.6	-2.2	19.1	33.2	4.3	0.4
30	250	4.7	7.5	51.5	-2.0	18.7	28.5	4.7	0.3	
30	350	6.5	7.2	51.7	-2.3	18.9	23.3	5.1	0.2	
30	0	200	5.3	10.0	65.6	-2.5	30.2	15.5	12.8	0.1
	0	300	5.0	13.4	67.6	-3.5	29.8	13.9	10.8	0.1
	10	200	5.4	14.7	53.7	-0.3	22.1	15.6	7.3	0.2
	10	300	3.3	9.9	55.5	-1.1	23.1	12.4	8.8	0.2
	20	200	5.5	13.3	54.4	-1.5	21.9	15.4	6.9	0.2
	20	300	5.3	17.9	57.4	-2.2	21.7	12.4	7.8	0.2
	30	200	7.1	6.3	49.5	-0.5	20.1	15.8	5.3	0.3
	30	300	6.3	8.8	49.3	-0.7	19.7	12.3	5.3	0.3
50	0	150	5.5	11.1	67.0	-1.6	31.1	18.0	13.4	0.1
	0	250	5.4	16.4	69.2	-3.2	30.0	13.4	12.0	0.1
	0	350	5.7	25.8	69.8	-4.4	29.3	11.7	8.8	0.1
	10	150	5.4	13.2	55.6	-1.6	22.1	18.0	7.3	0.2
	10	250	5.5	27.1	59.9	-2.7	22.9	13.1	7.3	0.2
	10	350	5.3	31.6	62.6	-3.8	22.8	10.7	6.3	0.1
	20	150	5.5	12.0	52.2	-1.2	20.6	14.2	6.3	0.3
	20	250	5.4	13.4	51.5	-1.0	20.6	13.6	6.0	0.2
	20	350	5.2	19.5	53.6	-1.7	21.0	10.5	6.3	0.2
	30	150	6.9	6.0	48.0	-1.1	18.6	17.9	3.8	0.5
	30	250	5.7	10.5	47.9	-1.2	18.4	13.8	4.0	0.4
	30	350	5.4	13.0	48.0	-1.2	18.8	12.1	4.3	0.3
70	0	200	6.2	20.3	69.8	-3.4	30.4	13.6	11.1	0.1
	10	200	6.0	17.3	54.6	-1.5	21.3	10.8	7.8	0.2
	10	300	5.6	17.0	58.0	-2.8	22.9	11.9	6.7	0.2
	20	200	5.4	16.4	53.3	-1.5	20.7	14.4	5.3	0.2
	20	300	5.3	21.1	54.5	-1.8	21.0	8.4	4.3	0.2
	30	200	5.6	13.1	48.7	-1.0	19.2	13.4	9.0	0.2
	30	300	5.5	16.9	48.5	-1.2	18.7	9.3	6.7	0.2
100	20	150	6.1	16.7	53.0	-0.3	22.5	12.0	6.0	0.2
	20	250	5.4	22.6	55.3	-0.7	22.5	7.3	5.6	0.2
	30	150	5.4	15.6	48.3	0.2	19.9	12.0	5.8	0.2
	30	250	5.3	21.4	50.0	0.1	20.2	9.4	5.3	0.2
	30	350	5.1	25.3	52.8	-0.6	20.5	5.9	5.1	0.2

WAI, water absorption index; WSI, water solubility index; SEI, sectional expansion index.

3.7. Image analysis

The image analyzing of extrudates at different levels of BSG and different screw speeds using a fixed formulation of starch (50%) and wheat flour (50%) were carried out (Figs. 1 and 2). Screw speed and BSG determined the extrudates internal structure, which influenced the textural properties of the final product. The growth and stability of

cells, their size, distribution, growth and failure during extrusion process have a major impact on the final quality of extrudates in terms of appearance and expansion depending on the organization of the cells in the extrudates. Image analysis of cross sections of extrudates showed that added starch increased the size of cells (data not shown). The visual image analysis in this study showed that the surfaces of a cross section of the extrudates appear

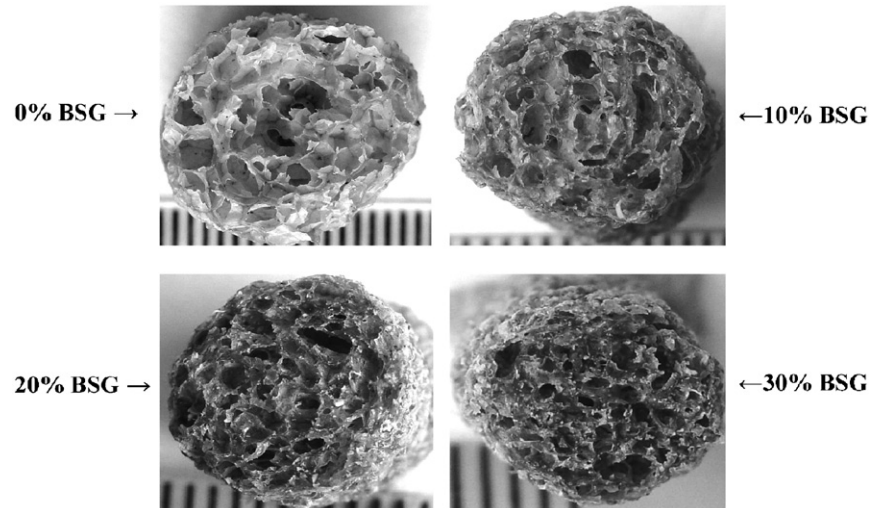


Fig. 1. Comparison of the cross section of extrudates containing different levels of BSG at a fixed level of starch (50%) and screw speed (150 rpm).

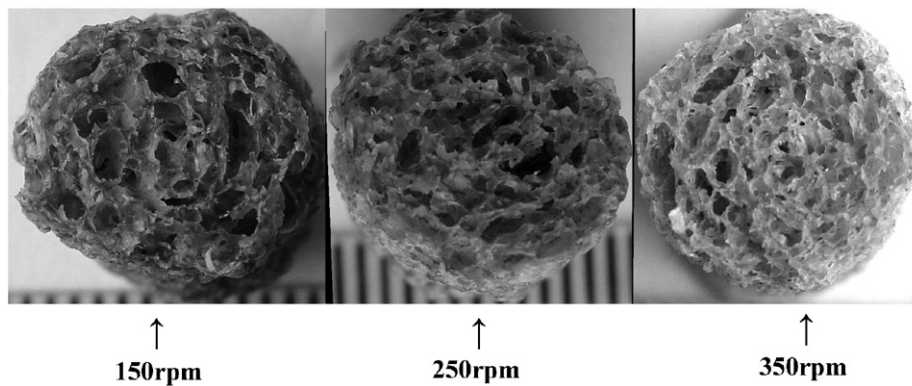


Fig. 2. Comparison of cross section of extrudates at different screw speed and fixed level of starch (50%) and BSG (10%).

to have a distribution of cell size. Images of extrudates showing the effect of different screw speeds and levels of BSG on the cell structure are presented in Figs. 1 and 2 and the corresponding dimensions of mean cells area are given in Table 5. Addition of BSG decreased mean cell area. Fig. 1 illustrates the cross section of extrudates containing different levels of BSG at a constant screw speed of 150 rpm. The total area of the cells varied between 15.80% and 47.00% of the total area of the cross section while total cells area in extrudates obtained with 10% BSG and screw speeds between 150 and 350 rpm (Fig. 2) were found to vary between 12.18% and 17.73%. The interaction of both BSG and starch in the present work resulted in the formation of a network of interconnected gas cells and a porous structure. The overall examination showed that, there was significant correlation ($P < 0.005$) between level of BSG to mean cells area in the product ($r = -0.7$). Similar findings were reported by Yanniotis et al. (2007) where wheat fibre in starchy extruded products resulted in extrudates with smaller cell size and higher number of cells. Mean cell area was highly correlated ($r^2 = 0.94$) to the SEI (Fig. 3)

Table 5
Mean cell area of the cross-section of the extrudates obtained at different levels of BSG and different screw speeds

BSG (g/100 g)	Screw speed (rpm)	Mean cell area (mm ²)
0	150	2.5
	250	2.2
	350	0.9
10	150	0.9
	250	0.7
	350	0.6
20	150	0.7
	250	0.7
	350	0.6
30	150	0.3
	250	0.4
	350	0.3

indicating that the higher level of BSG lowered the expansion of the product, giving a structure containing more small cells.

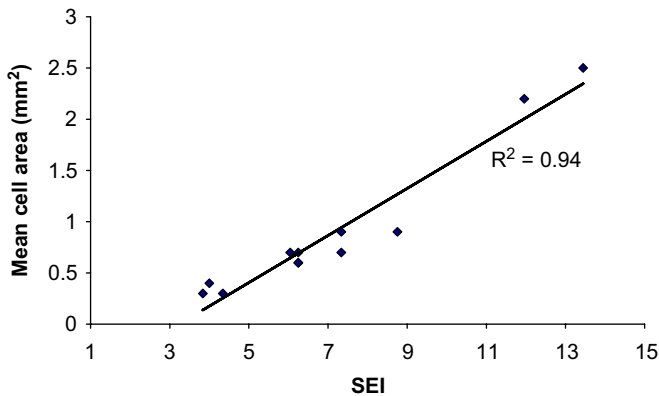


Fig. 3. Correlation between mean cell area and SEI.

4. Conclusion

This study shows that BSG can be successfully incorporated into ready-to-eat snacks. BSG addition can be used to improve the nutritional value of extrudates: significantly improving the dietary fibre level fat level and increasing crude proteins. Increasing the level of BSG, at levels between 10% and 30%, was found to proportionately reduce cell size, reduce the expansion of the product, increase the level of PA and increase the BD. Very little effect was observed with colour. Hardness of the product was reduced by increasing the level of starch. SME was affected by different screw speeds. With the incorporation of starch and the control of extrusion parameters, an acceptable expanded ready-to-eat snack can be produced. Up to 30% of BSG can be added with acceptable physicochemical characteristics, although the addition of 20% BSG was considered better for developing snacks with similar properties to those commercially available.

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