Sains Malaysiana 41(12)(2012): 1565–1570

Effect of Partial Replacement of Wheat Flour with Oyster Mushroom (*Pleurotus sajor-caju*) Powder on Nutritional Composition and Sensory Properties of Butter Biscuit

(Kesan Penggantian Sebahagian Tepung Gandum dengan Serbuk Cendawan Tiram Kelabu (*Pleurotus sajor-caju*) terhadap Komposisi Pemakanan dan Ciri-ciri Sensori Biskut Mentega)

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ABSTRACT

*Nutrient composition and sensory investigation of butter biscuits incorporated with various levels of grey oyster mushroom (*Pleurotus sajor-caju*, PSC) powder were studied. The biscuits were formulated with 0, 2, 4 or 6% of PSC powder to partially replace wheat flour. Our results showed that butter biscuit formulated with 6% PSC powder significantly ($p \leq 0.05$) recorded the highest protein content (6.94%) as compared with control (6.50%). Meanwhile, the addition of 2% PSC powder in biscuit does not affect the fat content (20.71%) compared with control (20.52%). Both biscuits containing 4% and 6% PSC powder recorded 5.34 g/ 100 g and 5.52 g/ 100 g total dietary fibre (TDF), respectively and significantly higher ($p \leq 0.05$) than control biscuit (4.84 g/ 100 g). Biscuits formulated with 4% and 6% PSC powder had significantly higher β-glucan concentration at 0.72 and 0.79 g/ 100 g than control biscuit (0.66 g/ 100 g). In the sensory evaluation, biscuits incorporated with 2% PSC powder had the highest scores for all sensory attributes except for crispiness and flavor. Butter biscuit containing 4% PSC powder had score values of 5.5 and 4.5 for crispiness and flavor, respectively. The present study suggested that incorporation of PSC powder up to 4% to replace wheat flour improved crispiness and flavour, increased concentration of protein, dietary fibre and β-glucan but did not affect the fat content of butter biscuit.*

*Keywords: Butter biscuit; nutrient composition; oyster mushroom (*Pleurotus sajor-caju PSC*); sensory evaluation*

ABSTRAK

*Komposisi nutrien dan kajian berkaitan penilaian sensori biskut mentega yang ditambah dengan cendawan tiram kelabu (*Pleurotus sajor-caju, PSC*) pada peratusan berbeza telah dikaji. Biskut mentega telah diformulasikan dengan serbuk PSC pada peratusan 0, 2, 4 atau 6% bagi menggantikan sebahagian tepung gandum. Keputusan kajian mendapati biskut cendawan yang diformulasikan dengan 6% serbuk PSC merekodkan kandungan protein yang paling tinggi dan signifikan ($p \leq 0.05$) iaitu 6.94% berbanding dengan kawalan (6.50%). Sementara itu, penambahan PSC pada paras 2% dalam biskut tidak mempengaruhi kandungan lemak (20.71%) berbanding dengan biskut kawalan (20.52%). Kedua-dua biskut yang mengandungi 4% dan 6% serbuk PSC merekodkan 5.34 g/ 100 g dan 5.52 g/ 100 g serat diet jumlah (TDF) dan signifikan ($p \leq 0.05$) berbanding biskut kawalan (4.84 g/ 100 g). Biskut yang diformulasikan dengan 4% dan 6% serbuk PSC mengandungi β-glukan yang tinggi dan signifikan iaitu sebanyak 0.72 dan 0.79 g/ 100 g berbanding dengan biskut kawalan (0.66 g/ 100 g). Penilaian sensori menunjukkan biskut yang ditambah dengan 2% PSC diberi skor yang tertinggi bagi semua atribut kecuali kerangupan dan perisa. Biskut mentega yang mengandungi 4% serbuk PSC merekodkan skor kerangupan dan perisa yang paling tinggi iaitu masing-masing 5.5 dan 4.5. Kajian ini menunjukkan penambahan serbuk PSC sehingga 4% bagi menggantikan sebahagian tepung gandum dapat memperbaiki kerangupan dan perisa atribut sensori, meningkatkan kandungan protein, serat diet dan β-glukan tetapi tidak mempengaruhi kandungan lemak biskut tersebut.*

*Kata kunci: Biskut mentega; cendawan tiram (*Pleurotus sajor-caju PSC*); komposisi makanan; penilaian sensori*

INTRODUCTION

Edible mushrooms are widely consumed as an important food item for their significant role in human health, nutrition and disease. Historically, mushrooms have been used for both medicinal and culinary properties in Asia and many parts of the world. Mushrooms have also been associated with many pharmacological properties by both eastern and western medicine. The functions include reducing cholesterol (Bobek et al. 1997), lowering blood pressure, strengthening the immune system against diseases (Reguła & Siwulski 2007), combating tumors (Mau et al. 2004) and improving liver function (Wang et al. 2000). Previous experimental evidence suggests oyster mushroom (*Pleurotus sajor-caju* PSC) inhibits hypertensive effects through its active ingredients, which affect the renin-angiotensin system (Chang 1996).

Oyster mushroom *(Pleurotus sajor-caju)* commonly grows on trunks and stumps of deciduous trees but presently it is also cultivated on various modified lignocellulosic substrates in shady cultivation farms. It possesses a very pleasant flavour and taste. The saccharidic complex of oyster mushroom is characterized by low content of digestible carbohydrates and by a relatively high content of polysaccharides which take part in formation of edible fibre (Strmiskova et al. 1992). Freshly harvested oyster mushrooms were reported to contain low fat content in average ranged from 0.38% to 2.28%, indicated low calorific value (kcal) contribution of mushrooms on total daily energy intake (Chye et al. 2008). On the other hand, the cultivated edible mushroom normally had high moisture content at more than 80%.

It contains up to 90% water, but owing to its protein and amino acids content, low fat and contain 9-group vitamins and a wide spectrum of mineral substances it represents a good source of biologically valuable substances for human nutrition (Strmiskova et al. 1992). Dry matters of mushrooms contain more than 25% protein, less than 3% crude fat and almost 50% of total carbohydrate (Kotwaliwale et al. 2007). Mushrooms are considered to be a healthy diet because it is low in calory, sodium, fat and cholesterol. Therefore, mushrooms form an important constituent of a diet for a population suffering from atherosclerosis (Dunkwal et al. 2007). It also contains appreciable amount of dietary fibre and β-glucan, vitamin B groups, D and other useful nutrients.

Bakery based products including biscuit are among top ten items popularly consumed by consumers in the Asia Pacific regions. Extensive studies have been done using various types of legumes, cereal and plant dietary fibre in bakery based products in attempting to enhance nutritional qualities and dietary fibre. The effect of utilization of legumes flour as a source of protein in bakery products (Eneche 1999; Hegazy & Faheid 1990; Patel & Rao 1995), cereal with milk or legumes flour (Adeyemi et al. 1989), defatted soy flour (Singh et al. 1996), chickpea and broad bean flours as well as isolated soy protein isolates (Rababah et al. 2006) has been reported previously. Literature review indicated that in general the nutritional, physical and sensory characteristic of biscuits depends on both the physicochemical properties of the legumes used in the formulation and on processing method employed for preparation of the legume flour. Recently, researchers found that dietary pigeon pea (*Cajanus cajan* L) grain has potential value as an economic source of protein and is widely consumed after appropriate processing (Tiwari et al. 2011) in biscuits. Another study found that by replacing wheat flour with lupin flour at 30% level in muffin formulation, substantial improvement in the protein and fibre contents can be achieved without affecting physical and sensory properties (Nieburg 2012).

It is expected that by partially replacing wheat flour with oyster mushroom powder into biscuit formulation, an improvement of nutritional composition especially dietary fibre without affecting sensorial properties can be achieved. Recently, we studied the colour, textural properties and cooking characteristics of chicken patty added with *Pleurotus sajor-caju* (PSC) (Wan Rosli et al. 2011). In continuation to this investigation, the incorporation of PSC in butter biscuit was conducted with the focus to enhance the nutritional composition and sensory qualities in bakery based products. This intention therefore necessitate that a thorough study be done to determine nutritional composition and sensory properties of butter biscuit added with PSC.

Materials and Methods

SAMPLE PREPARATION

Freshly harvested PSC was supplied by the National Kenaf and Tobacco Board of Malaysia (NKTB) from Bachok district of Kelantan, Malaysia. Fully-grown PSC with the pileus cap diameters between 9 and 11 cm were used throughout the study. The PSC was prepared by rinsing with clean water, blanched and chopped coarsely until the uniform sizes ranged from 2-5 mm is obtained. The chopped PSC was then oven dried (Mermert, Germany) at 50°C until constant weight was achieved. The dried PSC was then ground into powder form and sifted by using a siever having diameter of 125 μm. The PSC powder was kept in screw cap bottle at 4°C before further use.

BISCUIT PREPARATION

The biscuits were prepared by using common ingredients such as wheat flour, butter, table sugar, egg and leavening agent as shown in Table 1. PSC powder was partially substituted with wheat flour at the level of either 0 (control), 2, 4 or 6%. Composite flours and other dry ingredients (leavening agent and sugar) were mixed thoroughly in a mixing bowl before butter was added. Mixing was carried out in a Hobart mixer for 3-5 min to obtain creamy dough. The biscuit was then manually formed in a square shape with 5 mm of thickness and baked at 170°C for 15 min. The naked biscuits were then cooled at room temperature for 15 min before being kept in air tight polyethylene bags until further analyses.

PROXIMATE ANALYSES

Proximate analyses were conducted using AOAC (1996) for moisture, ash, insoluble dietary fibre (IDF), soluble dietary fibre (SDF) and total dietary fiber (TDF), protein by nitrogen conversion factor of 6.25 [Kjeldahl method, (AOAC 1996) and crude fat content using the semi-continuous extraction [Soxhlet] method (AOAC 1996). In this method, a homogenous ground sample (3 g for each) was dried in an oven at 105°C until constant weight was achieved. The difference between the initial weight and constant final weight after drying was considered as the moisture lost. Therefore, this difference was recorded as moisture content of the sample.

TABLE 1. Raw ingredients used in biscuit preparation

Ingredients (%)	PSC powder level (%)			
	0*	2	4	6
Composite flour				
(Wheat flour: PSC powder)	100:0	98:2	96:4	94:6
Butter	37	37	37	37
Egg	10.0	10.0	10.0	10.0
Sugar	24	24	24	24
Leavening agent	0.5	0.5	0.5	0.5

*The recipe was adopted from Brown (2008) with slight modification

Total ash content was determined by dry-ashing method, i.e. by incinerating a known quantity of dried food sample (0.5 g for each) in a muffle furnace at temperature 500-600°C until constant weight is obtained (AOAC 1996).

The semi-micro Kjeldahl method (AOAC 1996) was essentially used to determine the total nitrogen content of sample. The amount of nitrogen in the sample indicates the protein content of the sample after the amount of the nitrogen content is multiplied by a specific factor of 6.25 as most proteins contain 16% nitrogen. Exactly 1 g sample was oxidized by heating with nitrogen-free concentrated sulphuric acid in a long neck digestion flask, in the presence of selenium catalyst. In this digestion process, nitrogen in the sample was converted to ammonium sulphate. After that, the diluted digest was neutralized with concentrated sodium hydroxide solution (40%). Next, the ammonia was steam-distilled and trapped in saturated boric acid solution (4%). The volume of standard solution of hydrochloric acid used in titration will indicate the amount of ammonia liberated from the sample. Thus, the amount of nitrogen was determined.

Crude fat content was determined by direct extraction of dried ground sample (3 g for each) with petroleum ether in an intermittent extraction apparatus of the Soxhlet type. The residue in the extraction flask after the solvent was totally removed represented the fat content of the sample while the remaining in the extraction thimble became defatted sample. On the other nutrient, total carbohydrates were calculated by the difference: total carbohydrates = 100 – (g moisture + g protein + g fat + g ash). All measurements were carried out in triplicate (n = 3).

SENSORY EVALUATION

Sensory evaluations were carried out by 60 untrained consumers consisting of students and staffs of the School of Health Sciences, Universiti Sains Malaysia, Kubang Kerian. There were 18 male and 42 female Malays and Chinese with the age ranged from 20 to 38 involved in this study. Four biscuit samples with diameter approximately 3 cm × 0.5 cm thickness were presented to them for evaluation. The tested samples were coded with 3 digits permuted number. All samples were evaluated according to the 7-hedonic scaling method. They evaluated samples for aroma, colour, appearance, crispiness, flavour and overall acceptance on a 7 point scale (1 = dislike extremely and 7 = like extremely). Significance was established at $p \leq 0.05$ using statistics outline below.

STATISTICAL ANALYSIS

The data obtained were tested for significance using ANOVA and Duncan multiple range test with SPSS, Version 18. All measurements were conducted in triplicate (n=3). Significance level was established at $p \leq 0.05$.

RESULTS AND DISCUSSION

The nutritional composition of dried PSC is shown in Table 2. The PSC used in this study contained protein concentration of 23.3%. This value is close to the percentage range with those reported previously by Dikeman et al. (2005). They discovered that the protein content of various selected dried mushroom ranged from 23.4 to 43.5%. The fat concentration in oyster mushroom used in the present study is 3.0%. This value is close to the fat content of enokitake mushroom *(Flammulina velutipes)* which had 3.7% fat (Dikeman et al. 2005). The total ash content was recorded in oyster mushroom used in this study is 3.2%. Apart from that, dried oyster mushroom contained 35.6 g/ 100 g of TDF with IDF being the highest component (35.4 g/ 100 g) while SDF had the lowest value (0.2 g/ 100 g). The present results were in agreement with the dietary fibre content of the fruiting body of other mushroom species which ranged from 30-40% dry weight (Kurasawa et al.1982; Oyetayo et al. 2007). In addition other study revealed that other mushroom species such as *Poria cocos* also contained the dietary fiber content in this range (Cheung 1997).

The nutritional composition of biscuits prepared from composite flour of wheat flour replaced partially with PSC powder at different ratios is shown in Table 3. The results showed that the incorporation of PSC powder at different ratios resulted in significant different proportions of protein, fat and ash. Moisture content varied from 0.29 to 1.39% for biscuits prepared with increasing amounts of PSC powder to partially replaced wheat flour. It is indicated that incorporation of higher ratio of PSC powder to replace wheat flour significantly reduced moisture content. Result of this study indicated that all biscuits formulated with PSC

TABLE 2. Chemical compositions of dried PSC

Chemical compositions*	Concentration (%)
Moisture	90.2 ± 0.3
Protein	23.3 ± 0.9
Fat	3.0 ± 0.6
Ash	3.2 ± 0.01
Soluble dietary fibre	0.2 (g/ 100 g)
Insoluble dietary fibre	35.4 (g/ 100 g)
Total dietary fibre	35.6 (g/ 100 g)

*The analysis was replicated thrice (n = 3)

TABLE 3. Proximate analyses of biscuit incorporated with PSC (n=60)

Proximate compositions (%)	PSC Level (%)			
	0 (control)	2	4	6
Protein	6.50 ± 0.36[b]	6.66 ± 0.42[ab]	6.83 ± 0.09[ab]	6.94 ± 0.14[a]
Fat	20.52 ± 0.11[a]	20.71 ± 0.46[a]	21.40 ± 0.44[ab]	21.63 ± 0.60[b]
Moisture	1.39 ± 0.12 [a]	1.29 ± 0.06 [ab]	0.96 ± 0.02[b]	0.29 ± 0.01[c]
Ash	0.70 ± 0.15[b]	0.81 ± 0.05[a]	0.85 ± 0.06[a]	0.88 ± 0.16[a]
Carbohydrate	70.89 ±0.61[a]	70.53±0.70[a]	69.96±0.65[a]	70.26±0.72[a]

[a-b] Mean values within the same row bearing different superscripts differ significantly (p<0.05)

powder recorded significantly lower (p≤0.05) moisture content ranging from 0.29-1.29% as compared with control biscuit (1.39%).

The protein content of biscuits prepared from composite flour of wheat and PSC powder ranged from 6.50 to 6.94%. The protein content increased proportionally with PSC level used to replace wheat flour in biscuit formulations. Butter biscuit containing 6% PSC level significantly recorded the highest protein content (6.94%). However, the addition of PSC powder up to 4% does not increase significantly (p≤0.05) the protein content. These findings are in agreement with previous studies conducted by Ory and Conkerton (1983) on supplementation of bakery foods with high protein peanut flour and Singh et al. (1996) on the incorporation of defatted soya bean flour for the preparation of biscuits. Eneche (1999) also reported an increase in protein content of biscuits by the incorporation of pigeon pea flour with millet flour. Our findings indicated that the protein content of the biscuits improved with the incorporation of PSC powder and in line with previous study. Tiwari et al. (2011) reported that protein content of biscuit added with pigeon pea (*Cajanus cajan* L) had crude protein content ranging from 6.20 – 8.00 g/ 100 g.

Other result showed that the addition of 2% and 4% PSC powder does not affect the fat content (20.7% and 21.4%) compared with control (20.5%). However, the addition of PSC powder up to 6% slightly increased the fat content (21.63%) in the finished biscuit product. This slight elevation of fat content may be due to the fact that the original PSC powder used in biscuit formulation contains 3.0% fat (Table 2). Meanwhile, even though total ash content was found to increase in line with the PSC level in biscuit formulations, it was not significant. All biscuits recorded total ash content ranging from 0.70% - 0.88%. Hooda and Jood (2005) reported an increase in the total ash content of biscuits by incorporation of fenugreek flour.

Carbohydrates were among predominant macronutrients and ranged from 69.9 – 70.8% (Table 3), respectively, in all PSC-based biscuits and control biscuit. The values were not significant (p>0.05) among all PSC-based biscuits and control. The present data are within the range of values of the previous works done by other scientist. Tiwari et al. (2011) have documented that total carbohydrates content of cooked biscuit added with pigeon flour and wheat flour ranged from 66.51 – 83.48 g/ 100 g.

The present study showed that all butter biscuits containing PSC powder had higher amount of total dietary fibres (TDF) concentration. Both biscuits containing 4% and 6% PSC powder recorded 5.34 g/ 100 g and 5.52 g/ 100 g TDF, respectively and were significantly higher (p>0.05) than control biscuit (4.84 g/ 100 g). However, the addition of PSC powder at 2% in butter biscuit formulation (5.14 g/ 100 g) increased the TDF content compared with control biscuit (4.84 g/ 100 g) but not significant (p>0.05). The present result was in agreement with the dietary fibre content of the biscuit formulated with pigeon pea byproducts (Narasimha et al. 2004) and fenugreek flour (Hooda & Jood 2005). In our earlier study, chicken patty containing 50% ground oyster mushroom had significantly (p≤0.05) higher TDF at 4.90 g/ 100 g while chicken without addition of ground oyster mushroom had the lowest value at 1.9 g/ 100 g (Wan Rosli et al. 2011).

Both biscuit formulated with 4% and 6% PSC powder had significantly ($p \leq 0.05$) higher β-glucan concentration at 0.72 and 0.79 g/ 100 g. Even though, biscuit containing 2% PSC powder recorded higher β-glucan (0.68 g/ 100 g), it was not significant ($p > 0.05$) compared with control biscuit (0.66 g/ 100 g). Earlier study showed that chicken patty containing 50% fresh oyster mushroom recorded higher value of β-glucan and was significantly different ($p \leq 0.05$) when compared with chicken patty without containing ground oyster mushroom (Wan Rosli et al. 2011).

Table 5 shows the sensory evaluation scores for butter biscuits incorporated with PSC powder. Apparently, the scores of all sensory attributes were from 4.07-5.38. The present sensory data showed that all butter biscuits formulated with 2% PSC powder were not significantly different ($p > 0.05$) compared to the control biscuit for all attributes. Among all PSC-based biscuit treatments, the biscuits incorporated with 2% PSC powder had the highest scores for all sensory attributes except for crispiness and flavor. Meanwhile, butter biscuit containing 4% PSC powder had 5.51 and 4.54 score values of crispiness and flavor. These values are the highest among treatments but was not significant ($p > 0.05$) compared to other treatments (2% and control). The result also showed that butter biscuit containing 2% PSC powder had the highest scores of overall acceptance (5.16) followed by biscuit containing 4% PSC powder (5.02) and control (4.70). These results indicated that consumers prefer 2% of PSC powder to be incorporated in butter biscuit. The present study also showed that PSC powder level at 4% is preferred to be added in butter biscuit if the intention of having palatable flavor and crispiness texture of the finished baked items. This data indicated that consumers accept the biscuit prepared with PSC powder up to 4% of wheat flour replacement.

CONCLUSION

The addition of PSC powder up to 6% to partially replacing wheat flour in butter biscuit formulation resulted in increment of protein content. Both biscuits containing 4% and 6% PSC powder recorded significantly higher ($p \leq 0.05$) concentration of TDF at 5.34 g/ 100 g and 5.52 g/ 100 g, while β-glucan at 0.72 and 0.79 g/ 100 g, respectively. Biscuits incorporated with 2% PSC powder had the highest scores for all sensory attributes except for crispiness and flavor. Meanwhile, butter biscuit containing 4% PSC powder had 5.51 and 4.54 score values of crispiness and flavor. The present study suggested that incorporation of PSC powder up to 4% to replace wheat flour improves crispiness and flavour sensory attributes, increases concentration of protein, dietary fibre and β-glucan but unchanging fat content of butter biscuit. In summary, the addition of PSC powder resulted in an increased in the nutritional compositions, while maintaining the sensory quality of butter biscuit so they are as acceptable to consumers as butter biscuit produced with PSC powder.

ACKNOWLEDGEMENTS

The authors appreciate the funding from Universiti Sains Malaysia Delivering Excellence APEX Budget 2011 (1002/PPSK/910314), Nik Fakurudin, Solihah,

TABLE 4. Total dietary fibre (TDF) and B-glucan contents of mushrom biscuits

Nutrient compositions (g /100 g)	PSC Level (%)			
	0 (control)	2	4	6
TDF	4.84 ± 0.36[b]	5.14 ± 0.42[ab]	5.34 ± 0.09[a]	5.52 ± 0.14[a]
β-glucan	0.66 ± 0.05[b]	0.68 ± 0.03 [ab]	0.72 ± 0.05 [a]	0.79 ± 0.05 [a]

[a-b] Mean values within the same column bearing different superscripts differ significantly ($p < 0.05$)

TABLE 5. Sensory attributes of mushroom biscuit as influenced by the addition of PSC powder ($n = 60$)

Sensory attributes	PSC Level (%)			
	0	2	4	6
Aroma	4.21 ± 1.37[a]	4.74 ± 1.47[a]	4.64 ± 1.317[a]	4.51 ± 1.362[a]
Colour	5.15 ± 1.35[a]	5.18 ± 1.22[a]	4.34 ± 1.44[b]	4.44 ± 1.56[b]
Appearance	4.56 ± 1.46[ab]	4.62 ± 1.17[a]	4.23 ± 1.33[ab]	4.07 ± 1.30[b]
Crispiness	4.79 ± 1.83[b]	5.38 ± 1.46[a]	5.51 ± 1.51[a]	4.67 ± 1.58[b]
Flavour	4.16 ± 1.56[a]	4.48 ± 1.61[a]	4.54 ± 1.75[a]	4.41 ± 1.36[a]
Overall acceptance	4.70 ± 1.37[a]	5.16 ± 1.25[a]	5.02 ± 1.48[a]	4.69 ± 1.22[a]

[a-b] Mean values within the same row bearing different superscripts differ significantly ($p < 0.05$)
(Score 1 = dislike extremely and score 7 = like extremely)

Shazwan, Fatariah, Raushan and other staffs of School of Health Sciences who participated in sensory evaluation of PSC-based butter biscuit products. The authors would also like to thank the National Kenaf and Tobacco Board of Malaysia (304/PPSK/6150118/L120) and Anjaad Manufacturing Industries for their technical assistance in the research.

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Received: 2 April 2012
Accepted: 21 July 2012