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Evaluation of chromium fortified- parboiled rice coated with herbal extracts: resistant starch and glycemic index

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Abstract

The burden of diabetes has been increasing globally, particularly in developing countries, included Indonesia. Diet of low glycemic index (GI) foods will assist the diabetics in preventing and controlling the diabetes mellitus. Some nutrients that have been proven to improve insulin sensitivity include Cr and polyphenol found in cinnamon, pandan and bay leaf. This research aimed to evaluate content of resistant starch, and glycemic index of Cr - fortified - parboiled rice (Cr-PR) coated with herbal extracts. Variety of unhulled rice and forticant used in the experiment were Ciherang and CrCl3, respectively. Three herbal extracts used were cinnamon bark powder, pandan leaf and bay leaf with concentrations of 1, 2, and 3% of Cr-PR, respectively. Resistant starch (RS) content was determined by enzymatic process through glucooxydase method. Testing of the GI was conducted on 18 non-diabetic volunteers. After an overnight fast, every 2 volunteers were asked to consume 1 type of cooked rice sample from the 9 products produced and afterwards, 3 volunteers who had overnight fasts each were asked to drink 250 ml of water containing 50g glucose (as a food reference). The number of samples eaten is equivalent to 50 g of glucose. RS content of Cr-PR coated with herbal extracts ranged between 8.27 – 8.84% (dry weight). Cr-PR coated with a herbal extract type of 3% had higher RS levels than the ones with herbal extracts of 6% and 9% ($P < 0.05$). The highest RS content (8.84%) was attained by the rice which was coated with 3% cinnamon extract of 3%. The rice's GI ranged 29 - 40. The lowest GI (29-30) was attained by the Cr-PR coated with cinnamon extract of 6-9% The low GI of Cr-PR may be more influenced by the potential of polyphenolic compounds in the herbal extract than its RS levels.

Introduction

The prevalence and incidence of diabetes mellitus has increased drastically in the new industrial countries and developing countries, including Indonesia. The prevalence of diabetes for all age-group worldwide was estimated to be 2.8% in 2000 and 4.4% in 2030. The total number of people with diabetes is projected to rise from 171 million in 2000 to 366 million in 2030. In that year, the number of diabetics in Indonesia is predicted to reach 21.3 million, an increase of 150 percent from 8.43 million people in 2000 (Wild *et al*., 2004). World Health Organization reported that more than 80% of diabetes deaths occur in low- and middle-income countries (WHO, 2014). Globally, at least one in 10 deaths among adults aged 35-64 years is attributable to diabetes, rising to a quarter of all deaths in some parts of the world (Roglic and Unwin, 2005). International Diabetes Federation (IDF) states that two individuals develop diabetes every 10 seconds and two individuals die of diabetes related conditions every 10 second worldwide (IDF, 2007). One strategy that can be applied to manage diabetes is to consume foods that raise blood sugar slowly, but can provide the satisfaction of satiety. The trick is to consume food products that have a low glycemic index (GI). Therefore, it is necessary to provide a source of carbohydrates, especially rice as a staple food, which has a low GI, contains functional components that can lower blood sugar, and preferred by consumers. Larsen *et al.* (2000) reported that traditional parboiled rice had GI value of 46 and consumed the rice significantly reduced the blood sugar profile in type 2 diabetics rather than consuming non-parboiled rice (GI 55). The low GI can be attributed to high levels of resistant starch.

Naturally-occurring compounds that have been shown to improve insulin sensitivity and blood sugar control include Cr (Chromium) and polyphenols which are found in cinnamon *(Cinnamomun cassia)*

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(Anderson, 2008). Signs of Cr deficiency have the same signs as the metabolic syndrome (high blood sugar, high triglycerides, low density lipoproteins, and hypertension) and Cr supplementation has been shown to improve all signs of the deficiency in human subjects (Cefalu and Hu, 2004). Similar results are also demonstrated by the action of polyphenols from cinnamon, bay leaf, and pandan leaf, that the herb may increase insulin sensitivity or blood sugar control in experiments in vitro, animal, and human (Khan *et al.*, 2003; Hlebowicz *et al.*, 2007; Anderson, 2008; Agustin, 2010; Prameswari and Widjanarko, 2014).

Yulianto *et al.* (2012) reported that soaking time of 2.5 hours with $CrCl_3$ concentration of 7.47 mg / L at the parboiling stage could produce Cr-PR which had GI of 36.33, total Cr of 0.56 mg / kg and resistant starch of 11.88%. Although the chromium-fortified parboiled rice is classified as a low glycemic index food (<55) and is suitable for diabetics, but the sensory test result is less favored (score of 2.73) by panelists. On the other hand, non-parboiled rice was actually more preferred by the panelist with score of 4. Thus, this research aimed to evaluate the content of starch resistant, and glycemic index of Cr-PR coated with herbal extract (cinnamon, bay leaf and pandan leaf).

Materials and Methods

Materials and equipments

The long grain rough rice, Ciherang variety, was used for the experiment. The rice was obtained from the Indonesian Center for Rice Research located at Sukamandi, West Jawa. The chemicals used for fortification was chromium ($CrCl_3$, Sigma-Aldrich). The herbs used for rice flavor enhancers are Pandan leaves (*Pandanus amaryllifolius* Roxb.) and bay leaves (*Syzygium polyanthum* [Wigh] obtained from farmers in Argomulyo village, Sedayu sub-district, Bantul district, Special Region of Yogyakarta, Indonesia and cinnamon bark powder *(Cinnamomun cassia)*, Cap Pohon Bambu produced by UD. Bambu Hutan, Semarang, Indonesia. The study was conducted at the Laboratory of Agricultural Product Processing, Faculty of Agro-Industry, University of Mercu Buana Yogyakarta.

Production of Chromium Fortified- Parboiled Rice (Cr-PR)

Unhulled Ciherang rice containing 13% water was cleaned from impurities, such as gravel and soil. Ten kilograms of paddy were washed and discarded impurities (husk, sand, paddy straw), then soaked in a 15 L solution of $CrCl_3$ with concentration of 7.47 mg/L at a temperature of 65°C for 2.5 hours. Next, draining and steaming for 25 minutes, and cooled 0°C for 6 hours, then dried. The drying was done by using a cabinet dryer (made by FTI-UMBY) at a temperature of 50°C. The dry Cr fortified-parboiled paddy is milled to produce Cr-PR.

Preparation of herbal extracts

Herbal (cinnamon, bay leaf, and pandan leaf) extracts were prepared by using the method of Al-Jamal and Rasheed (2010). Five hundred grams of each herbal ingredient is soaked in 1,500 ml of hot water (88°C) in a water bath (88°C) in a water bath (Kottermann D-3162 Uetze-Hanigsen, W-Germany) for 6 hours. Then filtered with Whatman paper No. 41, and the filtrate obtained was collected in a dark bottle and stored in the refrigerator (4°C). Subsequently, the filtrate was concentrated by rotary vacuum (Buchi Labortechnik AG CH-9230 Flawil Swtzerland) at a temperature of 60°C to obtain 75 ml of herbal extract.

Coating method

Cr-PR was coated with various concentrations and types of herbal extracts (cinnamon bark, bay leaf and pandan leaf) using the Laohakunjit and Kerdchoechuen (2007) method with little modification of replacing 5% rice starch with gum arabic of 30 g. The coating substance was prepared by mixing gum arabic (powder type 4687, Brataco) 30 g with 100 ml of distilled water, and 2 drops of tween 80. Furthermore, the mixture was refrigerated for 12 hours, then added with 30 g of sorbitol (D-sorbitol solution 70%, Sigma-Aldrich). Concentrations of herbal extracts used in this experiment were 3, 6, and 9% (3, 6, and 9 ml for 100 g of Cr-PR). Coating process was done by spraying the mixture solution of 50 g in Cr- PR, then dried using a cabinet dryer at 50°C until the moisture content of 12-13%.

Determination of resistant starch (RS) and glycemic index (GI)

Cr-PR coated with herbal extracts was then analyzed for resistant starch, and glycemic index evaluation. RS content was determined by enzymatic process through glucooxydase method (Englyst *et al.*, 1992). Testing of the GI was conducted on 18 non-diabetic volunteers. After an overnight fast (10 hours), every 2 volunteers were asked to consume 1 type of the cooked Cr-PR coated with herbal extract containing the equivalent of 50 g of glucose from the 9 products produced. Two hours after ingesting the food, blood samples were taken every 30 minutes to measure the glucose levels (blood glucose level measured at the minute 30th, 60th, 90th, and 120th).

Seven days later, 3 volunteers who had overnight fasts each were asked to drink 250 ml of water containing 50g glucose (as a food reference) and measured their blood sugar levels every 30 minutes for 2 hours as in volunteers who ate rice samples. Blood glucose levels at each sampling time were plotted on two axes, namely the time axis (X) and blood glucose level axis (Y). GI was determined by dividing the area under the curve between GI measured food with a reference food (glucose) multiplied by 100, which GI value for glucose as the reference food = 100 (Wolever *et al.*, 1991; Foster-Powel *et al.*, 2002).

Statistical analysis

Analysis of variance (ANOVA) and test of significance were performed using SPSS (Statistical Analysis System Software) version-19 with confidence level 0f 95% (significance level $P < 0.05$). The samples were randomized for all the analyses describe above.

Results and Discussion

Resistant starch content

RS content of Cr-PR coated with different types and concentrations of herbal extracts are shown in Table 1. RS content of Cr-PR coated with herbal extracts decreased as the addition of herbal extracts concentration increased. The highest RS content, in the amount of 8.84%, was attained by the rice which was coated with cinnamon extract of 3%. Increasing the concentration of herbal extracts did not actually form more resistant starch content. The results of previous research indicated that parboiled rice modified (with cooling treatment) ranged from 8.97 - 9.48%, whereas treatment with Cr fortification could increase rice RS to 9.57 - 11.99% (Yulianto *et al.*, 2012). Thus, it seems that the addition of herbal extract does not encourage the process of forming the RS. The same result was also reported by Wu *et al.* (2009) who examined the effects of tea polyphenols (TPLs) on the retrogradation of RS. The result, after 10 days of storage at 4oC, RS gel with 10%, 14%, or 20% TPLs had almost no recrystallization of the retrogradation. The overall results demonstrate that the marked inhibitory effect of TPLs on the retrogradation of RS. The opposite is presented by Barros *et al.* (2012) that only treatments with sorghum proanthocyanidin increased RS in high amylose starch (amylose content = 66.5%). *Sorghum proanthocyanidins* interact strongly with starch, decreasing starch digestibility. Because this study is only a coating of herbal extracts on the surface of Cr-PR, there is little chance of interaction between polyphenolic compounds and starch.

Table 1 . Resistant starch (%) of Cr-PR coated with different types and concentration of herbal extracts

Herbal extract types	Concentration of herbal extract		
	3 %	6 %	9 %
Cinnamon bark powder	8.84 ± 0.23^{g}	8.41 ± 0.16^{c}	8.27 ± 0.10^{a}
Pandan leaf	8.78 ± 0.21^{f}	8.53 ± 0.18^{e}	8.56 ± 0.19^{e}
Bay leaf	8.49 ± 0.17^{d}	8.34 ± 0.12^{b}	8.31 ± 11^{b}

The numbers followed by the notation of the same letters showed that there was no significant difference at the level of 0.05 ($P<0.05$).

In the parboiling process, there is no stage which specifically directs the starch retrogradation. Nevertheless, the process of retrogradation on parboiled rice may occur during its storage process. Gelatinized starch contains no crystalline regions. Yet under certain conditions of storage and temperature, the molecules in starch gel can reassociate into an ordered stucture which is called retrogradation. Retrogradation of amylose fraction occured fast while retrogradation of amylopectin fraction was slow (Tako and Hizukuri, 2000). Retrogradation due to amylose is non reversible at temperatures less than 100^{o}C (Fitzgerald, 2004) because amylose crystals melt only at above 100^{o}C. Reported by Guraya *et al.* (2001), cooling of debranched non-waxy starch at 1^{o}C for 12 hr without agitation decreased digestibility by 59%; with stirring digestibility decreased by 42% after 24 hr cooling. Freezing of debranched cooled waxy and non-waxy starch does not affect the decrease in digestibility. Additional cooling process in the parboiling of this research provide a greater chance occurrence of starch retrogradation. The retrogradated starch is a form of resistant starch which is classified as RS3. RS can be formed naturally or during the processing of foods rich in starch (Sajilata *et al.*, 2006).

Glycemic Index

The change of blood sugar levels of healthy volunteers when fasting and 2 (hours) after consuming a standard diet (50 g of glucose) and Cr-PR with coated herbal extracts are shown in Figure 1, 2 and 3.

Based on the area under the curve of each volunteer's blood sugar response, the glycemic index value of Cr-PR coated with herbal extracts was then calculated with glucose as the standard (Table 2). The addition of different types and levels of herbal extracts significantly decreased blood sugar levels when compared with control (glucose). From Table 2, it can be seen that the GIs of Cr-PR coated with

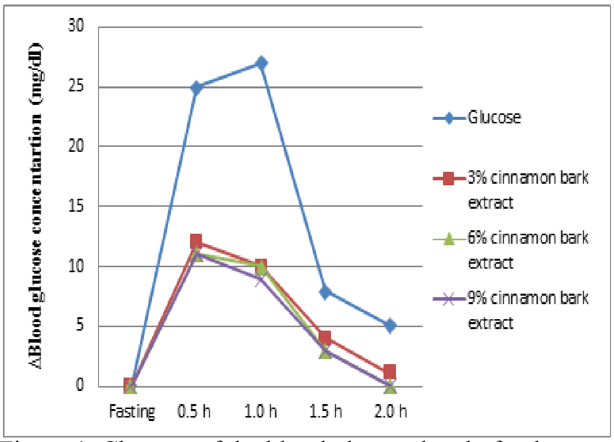

Figure 1. Changes of the blood glucose level of volunteers after consuming glucose (reference food) and cooked Cr-PR coated with cinnamon bark powder extract of 3%, 6%, and 9%.

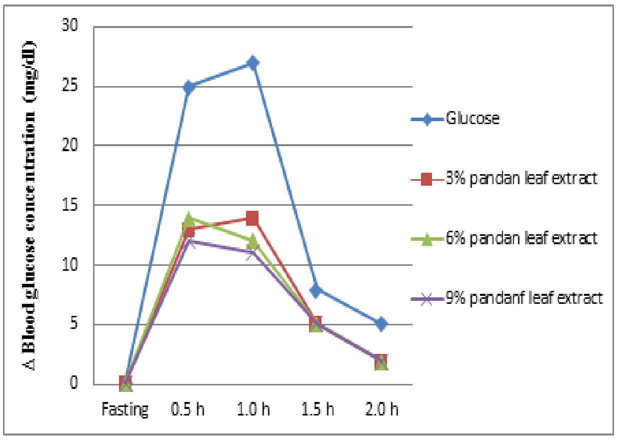

Figure 2. Changes of the blood glucose level of volunteers after consuming glucose (reference food) and cooked Cr-PR coated with pandan leaf extract of 3%, 6%, and 9%.

herbal extract of cinnamon, bay leaf and pandan leaf were low (29-40). Therefore, Cr-PR coated with enrichment of cinnamon, pandan and bay leaf extracts can be categorized as low GI food (<55).

The addition of cinnamon powder extract to 9% was able to decrease IG value (29), although it was not significantly different with the addition of cinnamon extract of 6% (IG 30). This low GI value is not supported by the magnitude of RS levels (Table 1), but can be caused by the high bioactive or phytochemical compounds contained in the cinnamon extract. Cinnamon herb was used not only because it is a common spices, but also it is known to be hypoglycemic, able to lessen blood sugar levels (Khan *et al.* , 2003). The compound in cinnamon which is believed to increase insulin receptor autophosphorylation and improve insulin sensitivity is prosianidin type-A polymers (Anderson *et al.* , 2004). These polyphenolic polymers found in cinnamon may function as antioxidants, potentiate insulin action, and may be beneficial in the control of glucose intolerance and diabetes. Reported by Dugoua *et al.* (2007) that two of three randomized clinical trials on diabetes type 2 sufferers provided strong scientific evidence that cassia cinnamon demonstrates a therapeutic effect in reducing fasting blood glucose by 10.3%-29%; the third clinical did not observe this effect. *Cinnamomum cassia*, however, did not have an effect of lowering glycosylated hemoglobin (HbA1c). Solomon and Blannin (2007) also reported that supplementation of cinnamon can control blood sugar and insulin sensitivity. Khan *et al.* (2003) reported as well that cinnamon intake of 1, 3 , or 6 g per day can lower serum blood sugar, triglycerides, LDL cholesterol, and total cholesterol of diabetes type 2 patients. These results indicate that cinnamon in the diet of diabetes type 2 patients will reduce risk factors concerning diabetes and cardiovascular diseases. Results were confirmed by Hlebowicz *et al.* (2007) which stated that intake of 6g of cinnamon with rice pudding decreases postprandial blood sugar (after taking it) and delayed gastric emptying. Preuss *et al.* (2006) also reported that cinnamon has a role in glucose metabolism and blood pressure regulation.

The addition of pandan leaves extract to 9% can significantly decrease IG value. The greater the pandanus leaf extract content added the lower the GI value of the produced rice (Table 2). The low GI value is not supported by the amount of RS (Table 1), but can be caused by the high bioactive or phytochemical compounds contained in pandanus extracts. Prameswari and Widjanarko (2014) reported that water extract of 'pandan wangi' leaves contained bioactive compounds such as tannins, alkaloids, flavonoids, and polyphenols with antioxidant activity of 66.82%. Water or ethanol extract of pandan leaf has hypoglycemic or antihyperglycemic activity. Chiabchalard and Nooron (2015) explained antihyperglycemic mechanism of pandan extract indicated that both water and ethanol extracts of *Pandanus amaryllifolius* Roxb. can inhibit the α-glucosidase enzyme and induce insulin production in mouse pancreatic cells (RINm5F).

The effect of addition of bay leaves extract is similar to the effect of the addition of pandan leaves extract; it's just the addition of bay leaves extract of 3% and 6% yield GI value which is not significantly different (Table 2). The addition of bay leaf extract to 9% can significantly decrease IG value. GI value of cooked Cr-PR coated with 9% bay leaves extract is lower than GI value of rice coated with 3% and 6% bay leaves extract. The low GI value is not supported by the amount of RS (Table 1), but can be caused by high bioactive or phytochemical compounds contained in

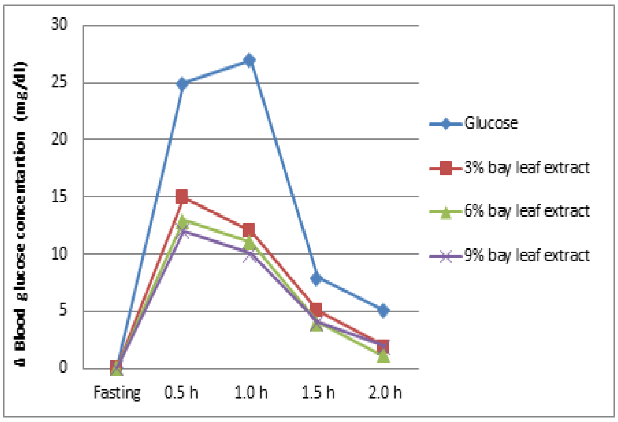

Figure 3. Changes of the blood glucose level of volunteers after consuming glucose (reference food) and cooked Cr-PR coated with bay leaf extract of 3%, 6%, and 9%.

Table 2. GI value of Cr-PR coated with different types and concentration of herbal extracts

Herbal extract types	Concentration of herbal extract		
	3%	6%	9%
Cinnamon bark powder	32.00 ± 2.05^{cd}	30.00 ± 2.16^{ab}	29.00 ± 2.04^{a}
Pandan leaf	40.00 ± 3.28^{f}	37.00 ± 2.55^{e}	33.00 ± 2.33^{d}
Bay leaf	37.00 ± 2.52^{e}	36.00 ± 2.40^{e}	31.00 ± 1.90^{bc}

The numbers followed by the notation of the same letters showed that there was no significant difference at the level of 0.05 (P<0.05).

the extract of bay leaf. Har and Ismali (2012) showed that the presence of gallic acid and caffeic acid as the major phenolic acids (antioxidant) in the methanolic *Syzygium polyanthum* leaves extract. Sukmadinata (2006) reported that addition of 15 ml of bay leaves extract into 200g of parboiled rice yields GI of 32 (cooked PR) and preferable cooked PR than control and one with bay leaves extract as much as 3 ml.

The GI value of parboiled rice is influenced by the processing factors. Starches cooked at high temperature for a long time and cooled can cause changes in the structure of the dissolved starch granules thereby encouraging the formation of retrogradated starch. This starch is insoluble and difficult to digest (resistant starch). RS levels of raw food are generally very low, but with processing and storage may increase levels of RS (Marsono, 1998). Parboiled rice fortified Cr of 7,47 mg / L and coated with 9% or 6% cinnamon herb extract had the lowest IG value (29-30) compared with other treatments. Although, rice of the results of this study generally can successfully lower blood sugar levels (GI value of rice is much lower than the control (glucose) .Thus, Cr-PR coated with extract of cinnamon, pandan and bay leaves can be used as an alternative diet food for diabetics, because it has low GI value or slowly increases blood sugar level.

Conclusion

The RS content of Cr-PR coated with herbal extract of cinnamon, pandan leaves, and bay leaves ranged from 8.27 to 8.84% (dry weight). Cr-PR coated with a herbal extract type of 3% had higher RS levels than the ones with herbal extracts of 6% and 9% (P <0.05). The highest RS level (8.84%) was achieved with Cr-PR coated with cinnamon extract of 3%. The GI value of Cr-PR coated with herbal extract ranged from 29 - 40. The lowest GI (29-30) was attained by the Cr-PR coated with cinnamon extract of 6-9%. The low GI of Cr-PR coated with herbal extracts may be more influenced by the potential of polyphenolic compounds in the herbal extract than its RS levels.

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