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Effect of Amaranth (*Amaranthus viridis*) Enriched White Bread Product on Nutritional Values and Antioxidant Capacity

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ABSTRACT

White bread is a popular food that is high in energy but low in other nutrients. This research aims to increase the nutritional value of white bread by using amaranth (*Amaranthus viridis*), a local plant with high nutritional value. Production of amaranth powder by blanching amaranth leaves at 80 °C for 2 min, then blending thoroughly and drying at 60 °C for 3 h. Amaranth powder supplementation in bread ranges from 0% (control), 2.5%, 5%, 7.5%, and 10% (w/w of flour weight). The results showed that bread fortified with 5% amaranth powder received the highest acceptance score, especially in the attributes of appearance, color, taste, and overall preference ($p \leq 0.05$). Fortification with amaranth powder significantly decreased L*, a*, and cohesiveness however it increased b*, hardness, springiness, and chewiness significantly compared to the control group. The addition of amaranth powder resulted in bread having higher nutritional values than the control, including protein, fat, ash, and crude fiber, while the carbohydrate content was reduced ($p \leq 0.05$). The antioxidant capacity of bread supplemented with amaranth powder by DPPH and ABTS assays was 8.34 and 2.71 TEAC (mg TE/g sample (dry basis)), respectively, and were greater than the control.

1. INTRODUCTION

Amaranth (*Amaranthus viridis*) is an herb with an upright, light green stem that is eaten as a vegetable in many parts of the world [1] including Thailand. Amaranth leaves and stems are good sources of proteins (about 35%), especially of essential amino acids for adults. It has been reported that 17 amino acids were detected in amaranth leaves from 20 standard amino acids that are components of proteins such as methionine, lysine, and asparagine [2]. In addition, it is also a source of dietary fiber and rich in minerals, i.e. magnesium, calcium, potassium, copper, phosphorus, zinc, iron, and manganese [2-4] and it has many potential pigments such as chlorophylls and carotenoids [5]. Moreover, amaranth leaves and seed extracts contain varied types of pharmacologically active compounds with antioxidant and antimicrobial activities such as vitamin C, flavonoids, phenolics, and tannins [2-5]. The results from several studies indicate that amaranth leaves have the potential to be used as a nutrient source to alleviate macro- and micronutrient deficiencies [7].

Several researchers reported the phytochemicals and antioxidants of amaranth leaves and stems. These natural antioxidants act as types of active oxygen (ROS) scavengers in the human body [8] and reactive nitrogen species produced as a result of oxidation are linked with different degenerative disorders such as cardiovascular diseases, cancer, cataracts, atherosclerosis, retinopathy, arthritis, emphysema, and neurodegenerative diseases [6,9].

White bread is generally considered less healthy than brown or whole wheat bread because it is made from refined flour, which has been stripped of its fiber, vitamins, and minerals during processing. Even though people generally know that whole wheat or whole grain bread has higher nutritional value. However, white bread is popular for consumption because it has a good taste, odor, and softness. Including, bread is one of the most versatile foods that can be personalized and fortified, responding to the needs of consumers from those with gluten intolerance to those with diabetes or cardiovascular disease. The addition of vegetable powders into bread can improve nutritional values such as fiber, polyphenol, antioxidant capacity, and rheological properties [10]. Therefore, this research aims to increase the nutritional value of white bread with amaranth, which are vegetable with high nutritional value that are easily available and inexpensive.

2. MATERIAL AND METHODS

2.1 Material and chemicals

The amaranth leaves were obtained from the local market. Other ingredients for the bread recipe were bread wheat flour, yeast, sugar, salt, milk powder, butter, and water. All chemicals used in the investigation were analytical grade.

2.2 Preparation of amaranth powder

The amaranth leaves were washed, cut into pieces, blanched at 80 °C for 2 min then dried at 60 °C for 3 h, ground into a fine powder, and stored in zipper bags for further use.

2.3 Preparation of white bread supplemented with amaranth powder

The ingredients used for making bread are shown in Table 1. Amaranth powder was added to white bread in the following percentages: 0% (control), 2.5%, 5%, 7.5%, and 10% (w/w of flour weight). The typical bread process involves mixing ingredients until the flour is converted into a dough, followed by baking the dough into a loaf.

Table 1. Five formulations of white bread supplemented with amaranth powder.

Ingredients	Quantities (g)				
	Control 0%	2.5%	5%	7.5%	10%
Bread flour	200	200	200	200	200
Yeast	1	1	1	1	1
Sugar	25	25	25	25	25
Salt	3	3	3	3	3
Milk powder	10	10	10	10	10
Butter	20	20	20	20	20
Water	130	130	130	130	130
Amaranth powder	-	5	10	15	20

2.4 Sensory evaluation

The bread of each sample was sliced 12 mm in thickness and ready to eat, without prior heating requirements. The sensory method was a 9-point hedonic scale with 30 untrained consumers (1= dislike extremely, 9= like extremely). The sensory aspects that were used in the evaluation were appearance, color, flavor, taste, texture, and overall preference.

2.5 Physicochemical properties analysis

Proximate composition i.e. moisture, protein, fat, ash, crude fiber, and carbohydrate was determined using the standard method of AOAC [11]. The color of bread samples was analyzed with a colorimeter (Hunter Lab), where L*, a*, and b* values were recorded. The texture profile analysis (TPA) of bread was evaluated by using a texture analyzer in the attributes of hardness, cohesiveness, springiness, and chewiness.

2.6 Antioxidant activity analysis

2.6.1 Extraction of bread for antioxidant activity analysis

The extraction of bread sample to determine the antioxidant activity including DPPH and ABTS assay. Briefly, 1 g of bread was ground and mixed with 1 ml of solvents acetone/water mixture (70:30 v/v); agitation with a vortex for 5 min at room temperature. Later on, the mixture was centrifuged at 800× g for 10 min [12]. Afterward, the supernatant was collected to analyze the antioxidant capacity following the procedure of Shimamura et al. and Sui et al. with some modifications [13,14].

2.6.2 DPPH radical scavenging activity

The DPPH (2,2-diphenyl-1-picrylhydrazyl) radical cation method was determined with the partial modification procedure of Shimamura et al. as follows: 100 µl of bread extract and 3.9 ml of DPPH solution (60 µM) were mixed in a cuvette and then incubated in the dark for 13 h at room temperature. Afterward, the absorbance of the mixture solution was measured at 515 nm using a UV-spectrophotometer. Trolox was used as the standard. The DPPH radical scavenging activity of bread was reported as Trolox equivalent antioxidant activity (TEAC) in mg Trolox equivalent (TE)/g sample (dry basis) [13].

2.6.3 ABTS radical scavenging activity

The ABTS (2,2'-azino-bis (3-ethylbenzthiazoline-6-sulphonic acid)) radical cation method was determined with the partial modification procedure of Sui et al. as follows: 100 µl of bread extract and diluted ABTS radical cation (ABTS•+) solution (3.9 mL) were mixed in a cuvette and then kept in the dark at room temperature for 30 min. Afterward, the absorbance of the mixture solution was measured at 734 nm using a UV spectrophotometer. Trolox was used as the standard. The ABTS value was reported as TEAC in mg TE/g sample (dry basis) [14].

2.7 Statistical analysis

The experiments were performed in triplicates and all results were expressed as mean ± standard deviation. The data were subjected to an analysis of variance (ANOVA) using SPSS. Significant differences among mean values were determined by Duncan's multiple range tests (DMRT), while the pairwise comparison of the mean using the Least significant difference (LSD) with a 95% confidence interval was significantly different ($p \leq 0.05$).

3 RESULTS AND DISCUSSION

3.1 *Sensory evaluation of bread*

The sensory evaluation results of bread supplemented with amaranth powder are shown in Table 2. Bread fortified with 5% amaranth powder showed significant differences in appearance, color, taste, and overall preference ($p \leq 0.05$). It was observed that when the amount of amaranth powder added to bread was higher than 5%, the liking scores of all attributes decreased with scores in the range of 5.04–7.11 (neither liked nor disliked to moderately liked). Therefore, 5% amaranth powder is the appropriate amount for adding to white bread.

Table 2. Sensory evaluation of white bread supplemented with different percentages of amaranth powder.

Sensory attribute	Amaranth powder substitution (%)				
	0	2.5	5	7.5	10
Appearance	7.12 ± 0.16^{b}	6.84 ± 0.07^{b}	7.46 ± 0.08^{a}	6.51 ± 0.15^{bc}	6.22 ± 0.28^{c}
Color	7.44 ± 0.05^{b}	7.42 ± 1.02^{b}	7.61 ± 0.05^{a}	7.02 ± 0.19^{c}	6.47 ± 0.08^{d}
Flavor	7.36 ± 1.15^{a}	7.34 ± 0.18^{a}	7.40 ± 1.01^{a}	6.87 ± 1.20^{b}	6.11 ± 0.06^{c}
Taste	7.43 ± 0.03^{b}	7.48 ± 0.38^{b}	7.77 ± 0.07^{a}	7.11 ± 1.08^{c}	6.72 ± 0.04^{d}
Texture	7.21 ± 1.03^{a}	7.17 ± 0.17^{a}	7.28 ± 0.11^{a}	6.50 ± 0.06^{b}	5.46 ± 0.05^{c}
overall preference	7.45 ± 0.15^{b}	7.34 ± 0.08^{b}	7.76 ± 0.08^{a}	6.53 ± 0.47^{c}	5.04 ± 0.09^{d}

Data are expressed as means ± SD values obtained from 30 untrained consumers. Different letters within the same row are significantly different at $p \leq 0.05$.

3.2 *Physicochemical properties of bread*

The appearance of white bread supplemented with different percentages of amaranth powder is shown in Figure 1. Different amounts of amaranth powder affected the appearance of bread. The color of the bread with amaranth powder added was green which is color-based of amaranth. In addition, the color of the bread gets darker when more amaranth powder is added.

Figure 1. White bread supplemented with different percentages of amaranth powder.

The result of the color and texture analysis of bread fortified with 5% amaranth powder compared with the control shown in Table 3. The addition of amaranth powder resulted in a decrease in the L* and a* values of the bread, while the b* value increased. Where the L* represents lightness from black to white on a scale of zero to 100, while a* and b* represent chromaticity with no specific numeric limits. Negative a* corresponds with green, positive a* corresponds with red, negative b* corresponds with blue and positive b* corresponds with yellow [15].

The texture profile analysis of bread fortified with 5% amaranth powder compared with control showed that the hardness, springiness, and chewiness increased when added amaranth powder, while cohesiveness decreased (Table 3). The amaranth bread had lower cohesiveness than

the control due to lower intermolecular force [10] while cohesiveness indicates the intensity of internal bonds of the structure that can be changed due to the addition of amaranth powder to the bread. While an increase in hardness was related to springiness and chewiness: hardness is the maximum force of food after compression that can represent the response of the bread during mastication compression between molars; springiness is the ability of a product to return to the original shape after initial compression that indicates the response of the structure when the bread is tested by hand compression; chewiness represents the energy needed for the destruction of a solid sample giving useful information on the consumer's perception during mastication.

Table 3. Color and texture profile analysis (TPA) of bread fortified with 5% amaranth powder compared with control.

Physical properties	Amaranth powder substitution (%)	
	0%	5%
L*	74.80 ± 1.68^{a}	59.18 ± 2.80^{b}
a*	8.36 ± 0.48^{a}	-7.83 ± 1.45^{b}
b*	25.28 ± 1.05^{b}	31.52 ± 5.16^{a}
Hardness (g)	59.33 ± 20.15^{b}	81.33 ± 46.73^{a}
Cohesiveness	0.97 ± 0.01^{a}	0.80 ± 0.64^{b}
Springiness (mm)	0.24 ± 0.15^{b}	0.35 ± 0.82^{a}
Chewiness (mJ)	10.64 ± 9.14^{b}	46.59 ± 37.09^{a}

Data are expressed as means ± SD value. Different letters within the same row are significantly different at p≤0.05.

The chemical composition of bread fortified with 5% amaranth powder compared with the control shown in Table 4. The addition of 5% amaranth powder increased the moisture, protein, fat, ash, and crude fiber content of bread (p≤0.05). Because the proximate composition of the amaranth leaves on a dry weight (DW) basis consists of 35% protein, 5.26 % fat, 21% ash, and 14% crude fiber [2].

Table 4. The chemical composition of bread fortified with 5% amaranth powder compared with the control.

Chemical composition (%)	Amaranth powder substitution (%)	
	0%	5%
Moisture	32.58 ± 0.07^{b}	34.57 ± 0.16^{a}
Protein	7.40 ± 0.23^{b}	9.98 ± 0.13^{a}
Fat	5.02 ± 0.05^{b}	6.10 ± 0.12^{a}
Ash	1.34 ± 0.02^{b}	1.62 ± 0.01^{a}
Crude fiber	0.00 ± 0.01^{b}	2.83 ± 0.12^{a}
Carbohydrate	53.65 ± 0.27^{a}	44.98 ± 0.06^{b}

Data are expressed as means ± SD values obtained from 3 replications. Different letters within the same row are significantly different at p≤0.05.

3.3 Antioxidant activity of bread

The antioxidant activity of bread is shown in Table 5. The DPPH and ABTS activity of amaranth powder before adding it to bread were 56.10 and 34.22 mg TE/g sample (dry basis) respectively. The bread fortified with 5% amaranth powder had significantly higher antioxidant capacity than the control both in the DPPH and ABTS assay (p ≤ 0.05).

Table 5. The antioxidant activity of bread fortified with 5% amaranth powder compared with the control.

Antioxidant activity TEAC (mg TE/ g sample (dry basis))	Amaranth powder substitution in bread (%)		Amaranth powder
	0%	5%	
DPPH	0.53 ± 0.01[b]	8.34 ± 0.06[a]	56.10 ± 0.02
ABTS	0.26 ± 0.21[b]	2.71 ± 0.02[a]	34.22 ± 0.01

Data are expressed as means ± SD values obtained from 3 replications. Different letters within the same row are significantly different at $p \leq 0.05$.

4 CONCLUSIONS

The finding of this research was used as a guide to design a recipe for bread or other food with the addition of amaranth powder. This is because amaranth leaves are a source of many beneficial nutrients. Bread recipes containing 5% amaranth powder can be used to develop functional breads because they are sensory accepted as well as adding nutritional value that is beneficial to the health of consumers.

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6 REFERENCES

[1] Grubb, A., & Raser-Rowland, A. (2012). The Weed Forager's Handbook. Hyland House Publishing Pty Ltd.

[2] Umar, K. J., Hassan, L. G., Dangoggo, S. M., Maigandi, S. A., & Sani, N. (2011). Nutritional and anti-nutritional profile of Spiny Amaranth (*Amaranthus viridis Linn*). Studia Universitatis Vasile Goldis Arad, Seria Stiintele Vietii, 21, 727–737.

[3] Sarker, U., & Oba, S. (2019). Nutraceuticals, antioxidant pigments, and phytochemicals in the leaves of Amaranthus spinosus and Amaranthus viridis weedy species. Scientific Reports, 9, 20413. https://doi.org/10.1038/s41598-019-50977-5

[4] Sarker, U., & Oba, S. (2019). Protein, dietary fiber, minerals, antioxidant pigments and phytochemicals, and antioxidant activity in selected red morph Amaranthus leafy vegetable. PLOS ONE, 14(12), e0222517. https://doi.org/10.1371/journal.pone.0222517

[5] Sarker, U., Hossain, Md. M., & Oba, S. (2020). Nutritional and antioxidant components and antioxidant capacity in green morph Amaranthus leafy vegetable. Scientific Reports, 10, 1336. https://doi.org/10.1038/s41598-020-57687-3

[6] Muhammad, J. I., Sumaira, H., Zahed, M., Farooq, A., & Amer, J. (2012). Antioxidant and antimicrobial activities of Chowlai (Amaranthus viridis L.) leaf and seed extracts. Journal of Medicinal Plants Research, 6(27), 4450–4455. https://doi.org/DOI: 10.5897/JMPR12.822

[7] Petras R. V., & Paulius, K. (2013). Nutritional components of amaranth seeds and vegetables: A review on composition, properties, and uses. Comprehensive Reviews in Food Science and Food Safety, 4, 381–412. https://doi.org/10.1111/1541-4337.12021

[8] Zaware, O. R., Wakchaure, R. S., Tambe, A. V., & Lokhande, R. P. (2022). Antioxidant activity of *Amaranthus Viridis Linn.* International Journal of Advanced Research in Science, Communication and Technology, 664–670. https://doi.org/10.48175/IJARSCT-4880

[9] Steffensen, S. K., Rinnan, A., Mortensenng, A. G., & Laursen, B. (2011). Variations in the polyphenol content of seeds of field-grown Amaranthus genotypes. Food Chem., 129, 131–138. https://doi.org/DOI:10.1016/j.foodchem.2011.04.044

[10] Tomosoiu, G. R., Cristea, S., Predescu, C. N., & Tudoreanu, L. (2022). Comparative study of textural properties of original bread containing vegetable powder and Romanian commercially available rye bread. Scientific Papers. Series B, Horticulture., 66(1), 932–937.

[11] AOAC (2005) Official method of Analysis (18th ed.). Association of Officiating Analytical Chemists, Washington DC.

[12] Blanco Canalis, M. S., Baroni, M. V., León, A. E., & Ribotta, P. D. (2020). Effect of peach puree incorporation on cookie quality and simulated digestion of polyphenols and antioxidant properties. Food Chemistry, 333. https://doi.org/10.1016/j.foodchem.2020.127464

[13] Shimamura, T., Sumikura, Y., Yamazaki, T., Tada, A., Kashiwagi, T., Ishikawa, H., Matsui, T., Sugimoto, N., Akiyama, H., & Ukeda, H. (2014). Applicability of the DPPH assay for evaluating the antioxidant capacity of food additives-Inter-laboratory evaluation study. Analytical Sciences: The International Journal of the Japan Society for Analytical Chemistry, 30(7), 717–721. https://doi.org/10.2116/analsci.30.717

[14] Sui, X., Dong, X., & Zhou, W. (2014). Combined effect of pH and high temperature on the stability and antioxidant capacity of two anthocyanins in aqueous solution. Food Chemistry, 163, 163–170. https://doi.org/10.1016/j.foodchem.2014.04.075

[15] What Is CIELAB Color Space? (n.d.). Retrieved May 5, 2024, from https://www.hunterlab.com/blog/what-is-cielab-color-space/