PJN
ISSN 1680-5194

PAKISTAN JOURNAL OF NUTRITION

ANSInet
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
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E-mail: editorpjn@gmail.com

OPEN ACCESS Pakistan Journal of Nutrition

ISSN 1680-5194
DOI: 10.3923/pjn.2019.587.594

Research Article

Natural Colorant from Black Rice Bran Improves Functional Properties and Consumer Acceptability of Yogurt

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Abstract

Background and Objective: Natural food colorants are utilized by the food industry as a safe health enhancing substitute for synthetic dyes. This study was conducted to investigate the effects of natural black rice bran colorant powder (BCP) addition on the properties of yogurt during storage at 4°C for 28. **Materials and Methods:** Plain yogurt and yogurt containing different levels of BCP [0.0125, 0.025, 0.05, 0.10 and 0.20% (%w/v)] were analyzed for total anthocyanins and phenolic contents, physicochemical quality parameters, functional properties and sensory characteristics. **Results:** Results showed that bioactive compounds and functional properties of yogurts with added BCP were improved and sensory characteristics were acceptable to panelists over storage time, with optimal level of BCP addition determined as 0.05%. Yogurts prepared by adding BCP contained higher total anthocyanins and exhibited higher liking scores in both color and appearance. **Conclusion:** Yogurt fortification with natural colorant powder from black rice bran has potential to improve both quality and functional properties of yogurt.

Key words: Anthocyanins, black rice bran, colorant powder, synthetic dyes, yogurt

Received: October 06, 2018 Accepted: March 13, 2019 Published: May 15, 2019

Citation: Patiwit Loypimai and Anuchita Moongngarm, 2019. Natural colorant from black rice bran improves functional properties and consumer acceptability of yogurt. Pak. J. Nutr., 18: 587-594.

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Yogurt is a dairy product obtained by fermentation of *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Streptococcus thermophilus* bacteria. Currently, yogurt containing fruits, sweeteners, flavoring and coloring agents has gained increasing consumer attention due to enhanced flavor and color balances[1,2]. Adding natural colorant from black rice bran, containing phytochemical compounds with probiotics as the main bioactive ingredient, to yogurt enhances health benefits and also improves consumer acceptability.

Black waxy rice (*Oryza sativa* L.) has recently gained popularity in terms of both consumption and use in the food industry because it contains high levels of phytochemicals which have health enhancing and disease prevention properties[3]. Among these compounds, dark purple anthocyanin pigments contribute both attractive color and strong antioxidant activity. The pigments are distributed mainly in the internal layers (pericarp and aleurone) of the bran[4]. In addition to these pigments, other bioactive compounds as polyphenols, γ-oryzanol and tocols (tocopherols and tocotrienols) are also present in the bran fraction[5-7]. Loypimai *et al.*[8,9] found that colorant powder prepared from black waxy rice bran by ohmic heating assisted extraction contained dark purple anthocyanins, tocols, carotenoids and γ-oryzanol. Loypimai *et al.*[8] reported that the majority of anthocyanins in black bran colorant were cyanidin-3-O-glucoside and showed a sevenfold increase compared with raw black bran. Application of this colorant offers a double positive benefit to foods by providing both functional substances and health effects while also improving appearance and enhancing consumer acceptability. BCP is, therefore, important to the food industry as a product containing both anthocyanin pigments and valuable bioactive compounds. In the present study, coloring agent from black rice bran was added to yogurt and physicochemical quality parameters, functional properties and sensory characteristics for consumer acceptability were investigated during cold storage at 4°C for 28 days.

MATERIALS AND METHODS

Preparation of colorant powder from black rice bran: Black waxy rice (*Oryza sativa var. glutinosa*.) bran sample and black bran colorant powder (BCP) were prepared following the procedure described by Loypimai *et al.*[8] and Duangmal *et al.*[10].

Preparation of yogurt: The model yogurt product was chosen because of its favorable low pH and brightly colored anthocyanins. Yogurt was produced according to the method described by Nontasan *et al.*[7] with slight modifications. Five different concentration levels of BCP [0.0125, 0.025, 0.05, 0.1 and 0.2% (% w/w)] were added to raw milk with BCP (0%) as control. All additional levels were acceptable under limitations of the Codex Alimentarius (no more than 0.2-3 mg kg^{-1} of milk product). Raw milk samples were pasteurized at 63°C for 30 min and then cooled to 45°C prior to adding a commercial bacterial starter. Samples were continuously incubated at 40-42°C until pH value reached 4.5-4.6 and then stored at 4°C until required for analysis.

Visual color measurement: Changes in visual color parameters were measured using the basic CIE L*, C* and h° color system of plain yogurt and yogurts added with BCP were measured using a Minolta Chroma Meter CR-300 (Konica Minolta, Japan) according to the method of Loypimai *et al.*[8].

pH and titratable acidity (TA)[11]: pH value change in plain yogurt and yogurts containing different levels of BCP were measured using a pH meter (Mettler Toledo, USA). Levels of total acidity in yogurts containing BCP and in plain yogurt (0% BCP) during storage at 4°C were determined using titration with 0.1 N NaOH. Results were calculated and expressed as %TA.

Determination of total anthocyanin content (TAC): Anthocyanin contents in both plain yogurt and yogurts with added BCP were analyzed according to the method described by Loypimai *et al.*[9] and Sarkis *et al.*[12] and Briefly, 25 mL of acidified methanol (methanol: HCl, 100:1 (v/v)) were added to 5.0 g of yogurt sample and then mixed and centrifuged at 6000×g (Rotina 48®, Universal 320R, Germany) for 5 min before total anthocyanin analysis.

TAC was performed by the pH differential method as described by Giusti and Wrolstad[13] and calculated in terms of Cy-3-glu (Loypimai *et al.*[9] for details).

Determination of total phenolic content (TPC): Extraction of TPC from each yogurt sample was performed according to the method of Singleton and Rossi[14] with slight modifications. The yogurt sample (2.0 g) was mixed with methanol-water (1: 1, v/v) and then extracted with a sonicator (50% amplitude, 130 W, 20 kHz, Vibra cell) for 5 min. TPC of the extracts was evaluated using Folin-Ciocalteu reagent according to the method of Iqbal *et al.*[15] A linear calibration curve of gallic acid standard was used to calculate TPC and the results were expressed as GAE g^{-1} sample.

Consumer acceptability: On the first day of storage, yogurt samples were evaluated for sensory acceptability by 40 untrained panelists who were all familiar with consuming yogurt products. The panelists consisted of staff and students of the Faculty of Science and Technology, Bansomdejchaopraya Rajabhat University, Bangkok, Thailand. Parameters measured were included color, flavor, taste, texture and overall acceptability of plain yogurt and yogurts added with five different levels of BCP using a nine-point hedonic scale (1 = very bad and 9 = excellent). The project was reviewed and approved by the Ethics Committee prior to beginning the experiment. Samples were presented monodically for each panelist. Appropriate level for BCP addition to the product was evaluated using a sensorial score (liking score) and compared with commercial yogurt served as the control sample (BCP, 0%).

Statistical analysis: Data were analyzed using one-way ANOVA with a completely randomized design (CRD) followed by Duncan's Multiple Range Tests using the Statistical Package for Social Science (SPSS) (SPSS Inc., Chicago, IL, USA). Significance was set at $p<0.05$.

RESULTS AND DISCUSSION

Changes in TA and pH value: Changes in total acidity and pH values of yogurts are shown in Fig. 1. Level of CP addition had no significant effect on total acidity and pH values of the product over storage time. Total acidity and pH values of plain yogurt and yogurts containing BCP ranged from 0.27-0.28% and 4.30-4.80, respectively. Thus, pH and total acidity values of yogurts added with BCP were comparable to the control sample (0% BCP). Moreover, pH values of all yogurts decreased progressively during storage for 28 days due to the lactic acid produced by yogurt bacteria. This result showed a similar trend to results reported by Obon *et al.*[16] and Nontasan *et al.*[7] According to the General Standard for Food Additives[17], use of coloring agent from plant materials as a food additive is technologically justified and authorized. Anthocyanin pigments in BCP are permitted for use as natural food colorant. Additionally, black rice bran is an edible fraction of the grain, prepared using an agri-solvent or a bio-solvent such as ethanol.

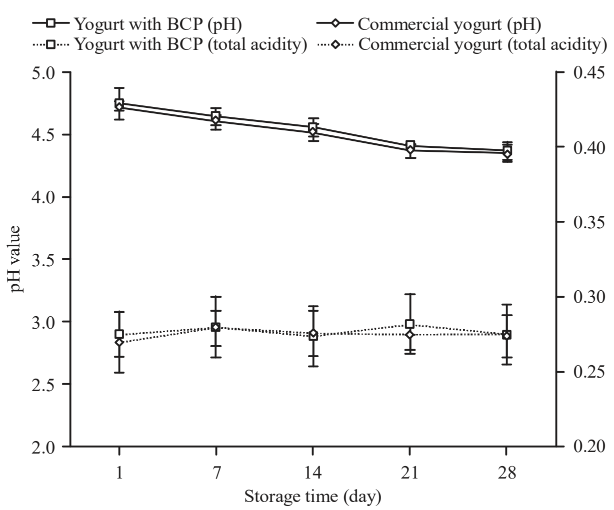

Fig. 1: Changes in total acidity and pH values of yogurts containing black bran colorant powder (BCP) and plain yogurt (0% BCP) during storage at 4°C

Changes in visual color: Color is a psychological property of food products affecting consumer acceptability. Visual color positively correlated with anthocyanin concentration in BCP[18]. Results of the present study indicated that level of additional BCP significantly influenced L*, C* and h° values of yogurts ($p<0.05$). C* value increased as level of BCP in yogurt increased, whereas L* and h° values decreased (Fig. 2). BCP level at 0.2% gave highest C* and lowest values of L* and h°. This may be due to the destruction of anthocyanin pigments during storage, as previously reported by Loypimai *et al.*[17]. Significant changes in C* were observed for all yogurts containing BCP. In current study, all yogurts containing BCP were purplish pink in color, depending on the additional levels which related to changes of anthocyanin content[17]. Degradation of total anthocyanins correlated positively with C* value, concurring with results of Nontasan *et al.*[7].

Total anthocyanin content (TAC): TACs of yogurts containing different levels of BCP are displayed in Table 1. Values showed significant difference between yogurts containing different levels of BCP and plain yogurt (0% BCP, control). Addition of BCP to yogurt significantly increased TAC. The yogurt sample with 0.20% BCP showed the greatest TAC because BCP contains high anthocyanin content, as reported in previous studies[8,9]. However, a slight increase of TAC in yogurt immediately after production was observed when BPC level increased. This may be due to degradation of anthocyanins in the yogurt by the pasteurization process. This result was similar to that observed by Loypimai *et al.*[17]; who found that anthocyanin pigments in BCP were susceptible to thermal processing, while He *et al.*[18] determined that preheated milk proteins increased the stability of anthocyanin extracts from grape skin. As shown in Table 1, all yogurts with BCP showed a decreasing trend over storage time, while plain yogurt with 0% BCP remained constant. Longer storage time promoted

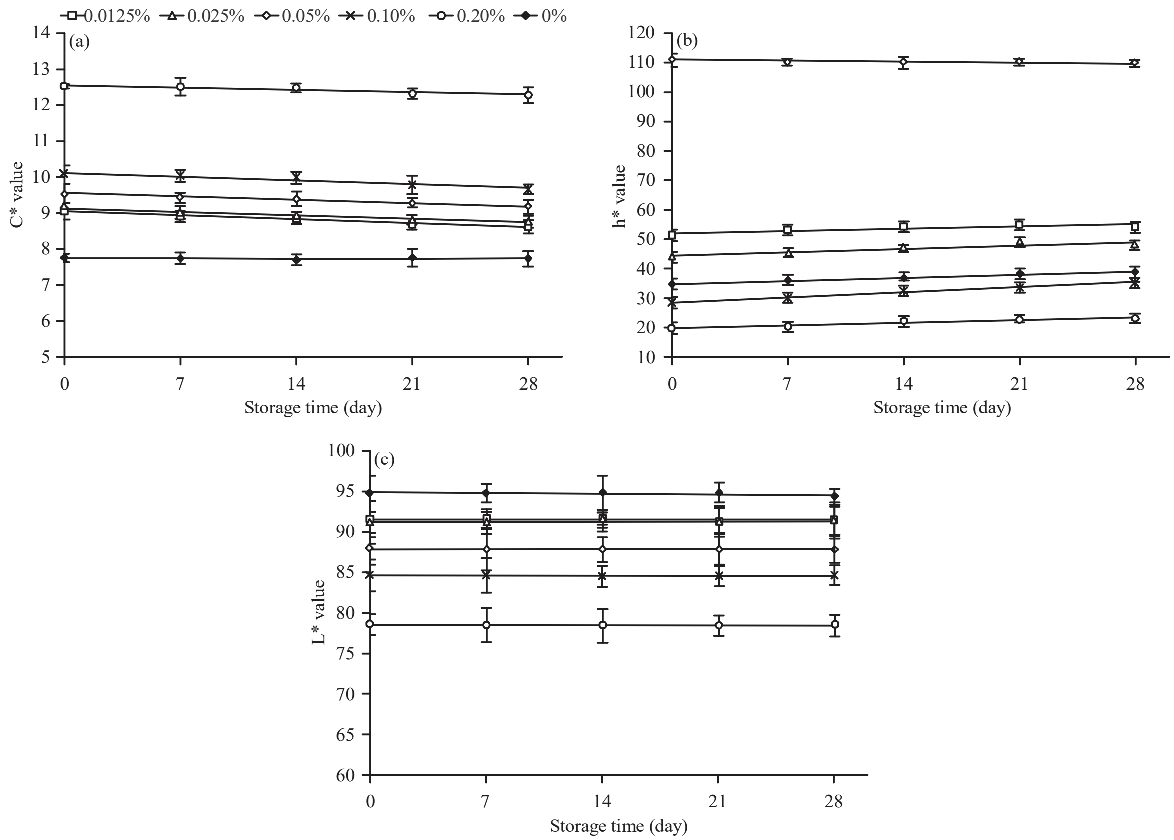

Fig. 2(a-c): Changes in visual color values of yogurts containing different levels of black bran colorant powder (BCP) and yogurt (0% BCP) during storage at 4°C

Table 1: Total anthocyanin and phenolic contents of plain yogurt and yogurts containing different levels of black bran colorant powder during storage at 4°C

	Storage time (day)			
BCP level (%)	0	7	14	21
Total anthocyanin content (mg/100 g)				
0.2	7.50±0.18^{A,a}	6.77±0.19^{A,b}	6.36±0.15^{A,c}	6.10±0.12^{A,d}
0.1	6.69±0.11^{B,a}	5.94±0.09^{B,b}	5.68±0.17^{B,c}	5.51±0.18^{B,d}
0.05	5.93±0.09^{C,a}	5.58±0.18^{C,b}	5.42±0.19^{C,c}	5.26±0.19^{C,d}
0.025	5.71±0.18^{D,a}	5.27±0.17^{D,b}	5.05±0.11^{D,c}	4.72±0.17^{D,d}
0.0125	4.25±0.15^{E,a}	3.77±0.19^{E,b}	3.52±0.09^{E,c}	3.16±0.15^{E,d}
0 (control)	nd	nd	nd	nd
Total phenolic content (µg GAE g^{-1})				
0.2	54.91±0.16^{A,a}	49.45±0.08^{A,b}	44.42±0.16^{A,c}	32.37±0.24^{A,d}
0.1	49.34±0.16^{B,a}	44.62±0.03^{B,b}	40.24±0.24^{B,c}	30.90±0.40^{B,d}
0.05	46.14±0.08^{C,a}	37.40±0.32^{C,b}	36.47±0.08^{C,c}	28.77±0.08^{C,d}
0.025	41.04±0.09^{D,a}	33.88±0.57^{D,b}	33.68±0.24^{D,c}	25.73±0.14^{D,d}
0.0125	38.27±0.11^{E,a}	31.34±1.63^{E,b}	29.91±0.24^{E,c}	23.19±0.08^{E,d}
0 (control)	2.16±0.24^{F,a}	1.84±0.08^{F,b}	1.23±0.08^{F,c}	0.71±0.05^{F,d}

Values are Means±SD of triplicate samples. nd: Not detected, Values with the same capital alphabet (A-F) in the same column are not significantly different ($p<0.05$). Values with the same small alphabet (a-d) along the same row are not significantly different ($p<0.05$), BCP: Black bran colorant powder, GAE: Gallic acid equivalent

the degradation of anthocyanin pigment. Similar results were found by Mozetic *et al.*[19] who reported that changes in visual color parameters such as C* strongly correlated with changes in anthocyanin concentration and were, therefore, considered as a good parameter to predict anthocyanin content[18]. In current study, a decrease in total anthocyanin content of all yogurt samples over storage time was observed and their color became duller. Stability of anthocyanins in yogurts is influenced by several conditions including storage (temperature, light), pH level, concentration of other phytochemicals (phenolic acids) and fat content[20] and also by properties of bacterial cultures used during milk fermentation. Previous studies Ozkan[21] and Yuksekdag *et al.*[22] also reported that hydrogen peroxide obtained from fermentation by lactic culture promoted the destruction of anthocyanins. Moreover, lactic cultures (i.e. *Lactobacillus paracasei* sub sp. *paracasei*) used in food production produce enzymes (i.e. ß-glucosidase) causing hydrolysis of anthocyanins to the less stable aglycone form[23-25] as the major reason for decrease in total anthocyanins during storage. Further investigation is required to determine the negative impacts of metabolic products, produced by yogurt bacterial starters, on the stability of BCP anthocyanins during yogurt storage.

Total phenolic content (TPC): TPC contents are shown in Table 1. All yogurt samples with added BCP had higher TPC values than the control (0% BCP). Yogurt with 0.2% BCP showed highest TPC content throughout storage time because BCP also contains phenolic compounds as well as anthocyanins, as reported previously by Loypimai *et al.*[9], whereas plain yogurt (0% BCP) is low in phenolic acids. Several bioactive compounds are present in milk, notably in fermented milk products; these are of great importance and include certain specific proteins, vitamins, bioactive peptides, organic acids and oligosaccharides[26]. Similar results were observed for yogurt products by Nontasan *et al.*[7], who reported higher values of polyphenolics obtained for natural black bran colorant powder than raw black bran. They observed a strong positive correlation between TPC and antioxidant activity assessed by FRAP assay.

Relationship between total anthocyanins and visual color: Correlations between anthocyanin content and visual color parameters namely L*, C* and h° values are presented in Fig. 3. The C* value decreased with decreasing anthocyanin content of yogurts containing BCP over storage time, whereas h° values increased. Anthocyanins occur in different chemical forms depending on the pH of the solution[17,27]. Anthocyanin content in sweet cherries (*Prunus avium* L.) had a positive correlation with the changes in C*[19]. Loypimai *et al.*[17] also pointed out that the C* value showed high correlation with cyanidin-3-O-glucoside (R^2>0.93) and total anthocyanins (R^2>0.92). These results concur with the results of Duangmal *et al.*[10] and Kara and Ercelebi[28].

Consumer acceptability: A maximum of 50% of non-dairy ingredients including sweeteners, flavors and other harmless harmless natural flavoring (coloring) ingredients can be mixed into fermented milk or yogurt[29]. Loypimai *et al.*[8,9] reported that BCP is considered as a natural functional colorant for application in flavored yogurt. In addition to analyses of physicochemical and functional properties, evaluation of consumer sensorial quality is also important. Sensorial assessment results are expressed as liking scores in Fig. 4. Liking scores of color, taste, appearance and overall acceptability of the yogurt samples were significantly

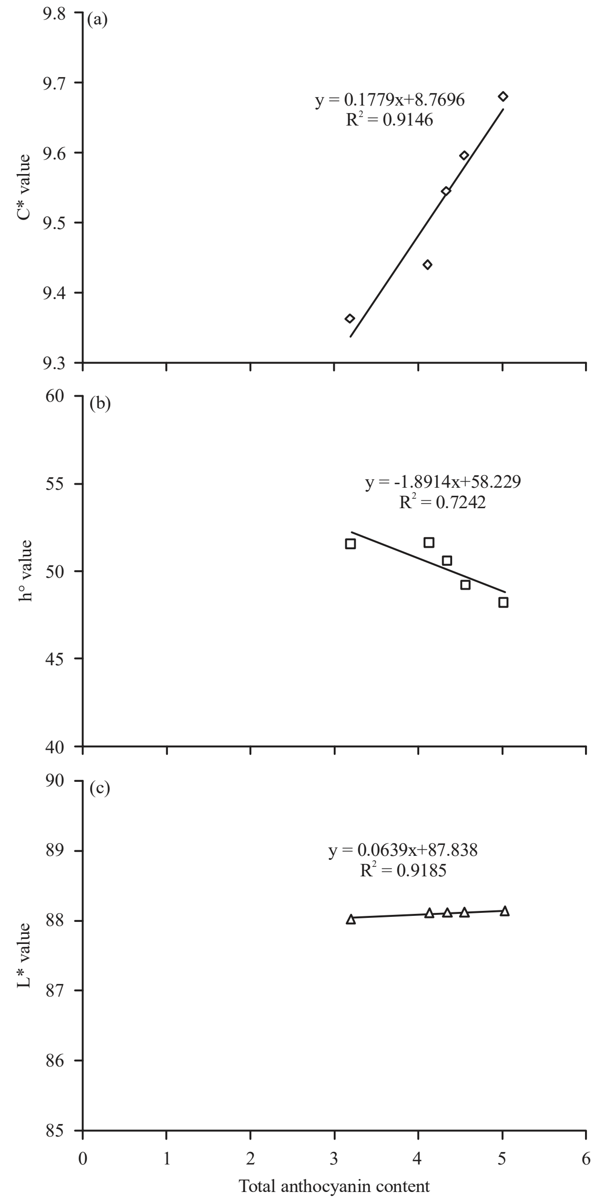

Fig. 3(a-c): Correlation between visual color (L*, C* and h° values) and anthocyanin content of yogurts containing black bran colorant powder (BCP) during storage

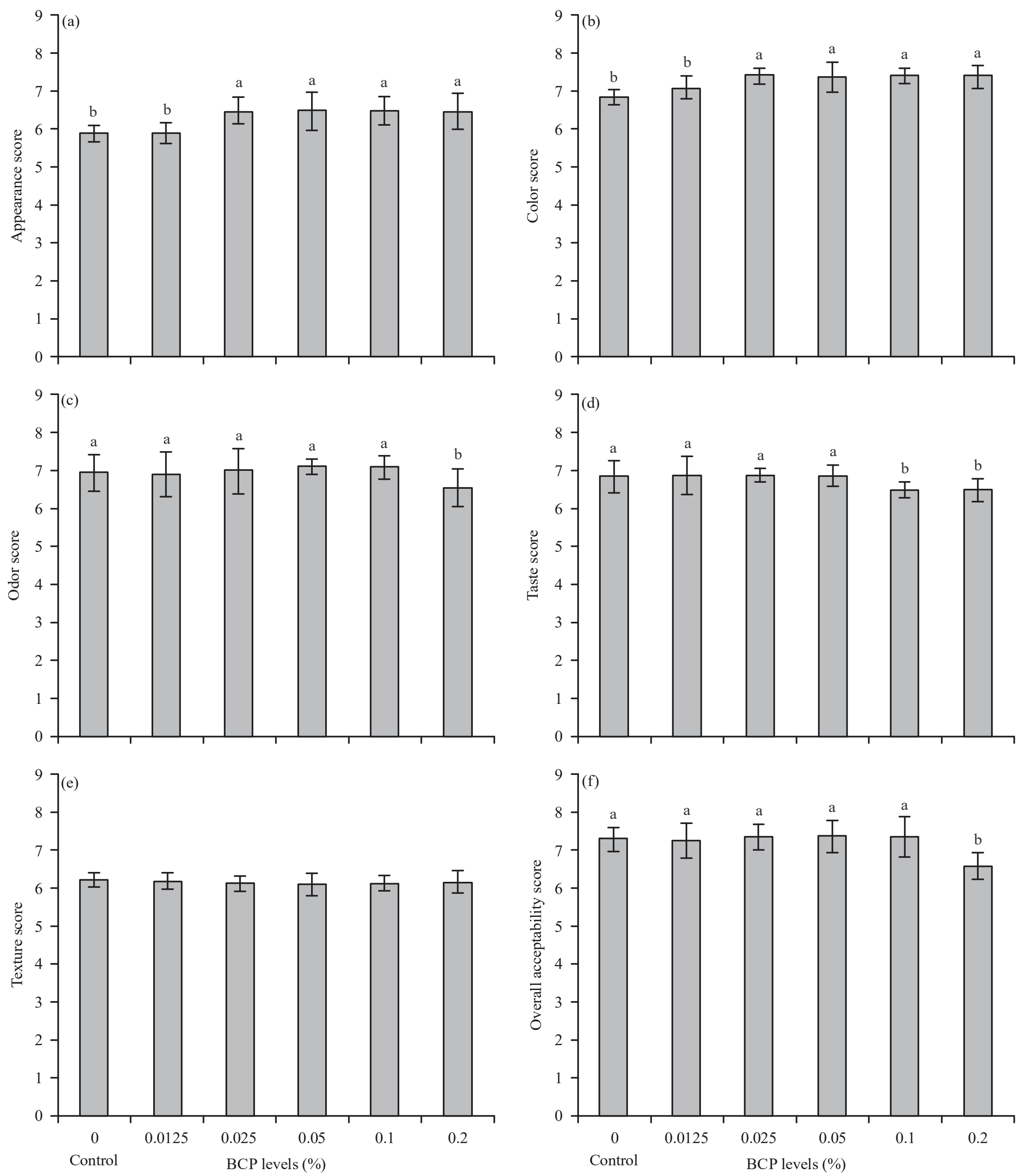

Fig. 4(a-f): Sensory quality scores of plain yogurt and yogurts containing different levels of BCP during storage at 4°C
The same letter above columns indicates no significant difference ($p<0.05$). Vertical bars on each column indicate standard deviation

different. Addition of BCP ranging from 0.025-0.2% obtained the best liking scores in color and appearance while liking scores in taste and odor decreased significantly in yogurts with more than 0.05 and 0.1% BCP, respectively. No significant difference was observed between yogurt samples in liking score for texture. Sensorial score in terms of overall acceptability was not significantly different among yogurts with no more than 0.1% BCP. Therefore, the optimal additional level was considered as 0.05% BCP, comparable with commercial yogurt (0% BCP) and sharing improved product color, appearance and functional properties.

CONCLUSION

Yogurts containing colorant powder prepared from black waxy rice bran could improve bioactive compounds of yogurt and consumer acceptability over storage time. Highest level of BCP addition was 0.05% with comparable acceptability of the panelists to the commercial product (0% BCP); however, the yogurt prepared by adding BCP exhibited higher liking scores in color and appearance and contained higher total anthocyanins. Results suggest that BCP has potential for use as a functional coloring agent for yogurt and also improves consumer acceptability.

SIGNIFICANCE STATEMENT

This study discovered the yogurt fortification with natural colorant powder from black rice bran which can improve the quality and functional properties of yogurt. This study will help the researchers to uncover the critical areas of application of black rice bran that many researchers were not able to explore. In this study, a new application of natural colorant powder from black rice bran was presented which improves consumer sensory satisfaction for yogurt.

ACKNOWLEDGMENTS

This research was financially supported by Mahasarakham University and Bansomdejchaopraya Rajabhat University, Thailand. The authors would like to thank Prof. Ian Warrington for his critical proofreading of the manuscript.

REFERENCES

1. Routray, W. and H.N. Mishra, 2011. Scientific and technical aspects of yogurt aroma and taste: A review. Comprehensive Rev. Food Sci. Food Saf., 10: 208-220.
2. Kiros, E., E. Seifu, G. Bultosa and W.K. Solomon, 2016. Effect of carrot juice and stabilizer on the physicochemical and microbiological properties of yoghurt. LWT-Food Sci. Technol., 693: 191-196.
3. Nam, S.H., S.P. Choi, M.Y. Kang, H.J. Koh, N. Kozukue and M. Friedman, 2006. Antioxidative activities of bran extracts from twenty one pigmented rice cultivars. Food Chem., 4: 613-620.
4. Yawadio, R., S. Tanimori and N. Morita, 2007. Identification of phenolic compounds isolated from pigmented rices and their aldose reductase inhibitory activities. Food Chem., 101: 1616-1625.
5. Qureshi, A.A., S.A. Sami, W.A. Salser and F.A. Khan, 2002. Dose-dependent suppression of serum cholesterol by tocotrienol-rich fraction (TRF_{25}) of rice bran in hypercholesterolemic humans. Atherosclerosis, 161: 199-207.
6. Loypimai, P., A. Moonggarm and P. Chottanom, 2009. Effects of ohmic heating on lipase activity, bioactive compounds and antioxidant activity of rice bran. Aust. J. Basic Applied Sci., 3: 3642-3652.
7. Nontasan, S., A. Moongngarm and S. Deeseenthum, 2012. Application of functional colorant prepared from black rice bran in yogurt. APCBEE Proc., 2: 62-67.
8. Loypimai, P., A. Moongngarm, P. Chottanom and T. Moontree, 2015. Ohmic heating-assisted extraction of anthocyanins from black rice bran to prepare a natural food colourant. Innovat. Food Sci. Emerg. Technol., 27: 102-110.
9. Loypimai, P., A. Moongngarm and P. Chottanom, 2016. Phytochemicals and antioxidant capacity of natural food colorant prepared from black waxy rice bran. Food Biosci., 15: 34-41.
10. Duangmal, K., B. Saicheua and S. Sueeprasan, 2008. Colour evaluation of freeze-dried roselle extract as a natural food colorant in a model system of a drink. LWT-Food Sci. Technol., 41: 1437-1445.
11. Fabro, M.A., H.V. Milanesio, L.M. Robert, J.L. Speranza, M. Murphy, G. Rodriguez and R. Castaneda, 2006. Technical note: Determination of acidity in whole raw milk: Comparison of results obtained by two different analytical methods. J. Dairy Sci., 89: 859-861.
12. Sarkis, J.R., D.P. Jaeschke, I.C. Tessaro and L.D. Marczak, 2013. Effects of ohmic and conventional heating on anthocyanin degradation during the processing of blueberry pulp. LWT-Food Sci. Technol., 51: 79-85.
13. Giusti, M.M. and R.E. Wrolstad, 1996. Characterization of red radish anthocyanins. J. Food Sci., 61: 322-326.
14. Singleton, V.L. and J.A. Rossi, 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Viticult., 16: 144-158.
15. Iqbal, S., M.I. Bhanger and F. Anwar, 2005. Antioxidant properties and components of some commercially available varieties of rice bran in Pakistan. Food Chem., 93: 265-272.
16. Obon, J.M., M.R. Castellar, M. Alacid and J.A. Fernandez-Lopez, 2009. Production of a red-purple food colorant from *Opuntia stricta* fruits by spray drying and its application in food model systems. J. Food Eng., 90: 471-479.
17. Loypimai, P., A. Moongngarm and P. Chottanom, 2016. Thermal and pH degradation kinetics of anthocyanins in natural food colorant prepared from black rice bran. J. Food Sci. Technol., 53: 461-470.
18. He, Z., M. Xu, M. Zeng, F. Qin and J. Chen, 2016. Preheated milk proteins improve the stability of grape skin anthocyanins extracts. Food Chem., 210: 221-227.

19. Mozetic, B., P. Trebse, M. Simcic and J. Hribar, 2004. Changes of anthocyanins and hydroxycinnamic acids affecting the skin colour during maturation of sweet cherries (*Prunus avium* L.). LWT-Food Sci. Technol., 37: 123-128.
20. Wallace, T.C. and M.M. Giusti, 2008. Determination of color, pigment and phenolic stability in yogurt systems colored with nonacylated anthocyanins from *Berberis boliviana* L. as compared to other natural/synthetic colorants. J. Food Sci., 73: C241-C248.
21. Ozkan, M., 2002. Degradation of anthocyanins in sour cherry and pomegranate juices by hydrogen peroxide in the presence of added ascorbic acid. Food Chem., 78: 499-504.
22. Yuksekdag, Z.N., Y. Beyath and B. Aslim, 2004. Metabolic activities of *Lactobacillus* spp. strains isolated from kefir. Food/Nahrung, 48: 218-220.
23. Buchert, J., J.M. Koponen, M. Suutarinen, A. Mustranta, M. Lille, R. Torronen and K. Poutanen, 2005. Effect of enzyme aided pressing on anthocyanin yield and profiles in bilberry and blackcurrant juices. J. Sci. Food Agric., 85: 2548-2556.
24. Otieno, D.O., J.F. Ashton and N.E. Shah, 2005. Stability of β glucosidase activity produced by *Bifidobacterium* and *Lactobacillus* spp. in fermented soymilk during processing and storage. J. Food Sci., 70: M236-M241.
25. Arnous, A. and A.S. Meyer, 2010. Discriminated release of phenolic substances from red wine grape skins (*Vitis vinifera* L.) by multicomponent enzymes treatment. Biochem. Eng. J., 49: 68-77.
26. Scibisz, I., M. Ziarno, M. Mitek and D. Zaręba, 2012. Effect of probiotic cultures on the stability of anthocyanins in blueberry yoghurts. LWT-Food Sci. Technol., 49: 208-212.
27. Castaneda-Ovando, A., M. de Lourdes Pacheco-Hernandez, M.E. Paez-Hernandez, J.A. Rodriguez and C.A. Galan-Vidal, 2009. Chemical studies of anthocyanins: A review. Food Chem., 113: 859-871.
28. Kara, S. and E.A. Ercelebi, 2013. Thermal degradation kinetics of anthocyanins and visual colour of Urmu mulberry (*Morus nigra* L.). J. Food Eng., 116: 541-547.
29. Codex Alimentarius, 2003. Codex standard for fermented milks. Codex Stan 243-2003. www.codexalimentarius.net/download/standards/400/CXS_243e.pdf