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Physicochemical, antioxidant capacity, and sensory properties of bligo juice (*Benicasa hispida*) with bligo peel extract addition

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Abstract

This study aimed to develop bligo juice as functional food by adding Bligo Peel Extract (BPE) with concentrations of 1%, 2%, and 3%. The characteristics of bligo juice before and after BPE addition were investigated by proximate analysis, examination of total solids, viscosity, vitamin C, and antioxidant capacity, and sensory evaluation. Results showed that BPE addition increased the vitamin C levels, antioxidant activity, carbohydrates, fats, and acceptance of color, flavor, and viscosity of bligo juice but decreased its acceptance of taste and acceptability. The addition of 1% BPE provided the highest acceptance rate compared with other variations. In summary, BPE addition increases the vitamin C, antioxidant activity, and carbohydrate content of bligo juice, thus improving its health benefits.

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1. Introduction

Bligo (*Benincasa hispida*), also known as *kundur*, winter melon, or ash gourd, is a member of Cucurbitacea family and is widely grown in Asian countries. After hours of boiling, This fruit is traditionally used as an ingredient in soups, salads, and most commonly in drinks (1). Bligo contains nutrients such as lipids, proteins, carbohydrates, fiber, minerals, and vitamins and has positive effect on diabetes, liver, and fever (2,3). Nadhiya *et al.* (4) showed that the ethanolic extract of bligo has high amounts of phenols and flavonoids and exhibits good scavenging capacity for superoxide anion radicals with IC_{50} of 271 ± 1.89 µg/ml, making it a potential antioxidant. Bligo has a high water content (93.80–96.80%), a low sugar content (1–2%, w/w), and a high dietary fiber level (27.5% of the dry weight), make it a potential candidate for low-calorie juice (1,5).

Researchers attempted to develop bligo as a functional drink. Alsuhendra *et al.* (6) prepared bligo juice by extracting bligo pulp and found the highest potassium content of 1.882 mg/kg. Aini (7) developed bligo juice with stabilizers (Carboxy Methyl Cellulose (CMC), pectin, and gum arabic) and sucrose (7, 8, and 9%) under different pasteurization temperatures (70, 80, and 90°C) and found that CMC provided the best response for stability, 70 °C pasteurization temperature resulted in the highest vitamin C content (60.167 Vit C/100 ml), and 9% sucrose addition was the most preferred by panelists in terms of taste and aroma.

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Sun *et al.* (8) reported the effects of filtration (pulp reduction) and thermal processing (boiling) on the quality of bligo juice. Phenolic amino acids phenylalanine, tyrosine, and tryptophan were detected at 10–45 mg/L, and the antioxidant activity ranged 36–49 mg gallic acid/L. The bligo juice was high in insoluble solids and antioxidants and low in sugar and acid.

In contrast to its pulp, bligo peel is rarely used and becomes a waste product with potential functional property. According to its nutritional content, bligo peel contains more protein (3.60 g/100g), calcium (692 mg/100 g), and iron (17.06 mg/100 g) than bligo pulp. Nagarajaiah and Prakash (9) compared the nutritional content of several Cucurbitacea peels including bligo, chayote (*Sechium edule*), and oyong (*Luffa acutangula*) and found that bligo peel has higher dietary fiber, iron, and calcium contents than chayote and oyong peels. Rana and Sutee (10) conducted phytochemical screening to investigate the antioxidant potential of bligo peel using DPPH and reported the values of 87.87% and 86.5% at 100 µg/mL for its aqueous and methanol extracts, respectively. Abdullah *et al*. (11) showed that the total phenol content of fruit peel is the smallest at 74.83 GAE/g compared with pulp and seeds. In our previous study, the antioxidant activity of bligo peel include in medium category (IC_{50} 6.91 mg/ml) (12). Kumar *et al.* (13) studied the sugar type and content in bligo peel using thin-layer chromatography and found galactose, glucose, xylose, and sorbose in the methanol extract of bligo peel.

Bligo juice has been added with various sugars and stabilizers (such as CMC) and subjected to different treatments to increase its physiochemical and functional properties. However, the use of Bligo Peel Extract (BPE) to increase the functional value of bligo juice has not been explored, except for Sun *et al*. (14) who prepared bligo juice without removing the peel but did not examine the effectiveness of bligo peel in enhancing functional properties. Therefore, the current research was conducted to investigate the effect of BPE addition on the physicochemical, antioxidant activity, and sensory attributes of bligo juice. The results provide new insights into increasing the value of local commodity and thus reducing waste.

2. Materials and Methods

2.1. Materials

Mature bligo fruits with an oval shape and length of 20–30 cm were obtained from Jepara, Central Java, Indonesia. Bligo peel was removed, dried for 5 days, and ground to powder. Extraction was performed by maceration. The sample was dissolved with aquadest solvent in a ratio of 1:6, placed in a room protected from light and then allowed to stand at room temperature, filtered using filter paper and a Buchner funnel, and finally dried using a rotary evaporator.

2.2. Research Procedures

Bligo juice was prepared using the method of Alsuhendra *et al*. (6) with modifications. Bligo fruit was cut, washed, cleaned, and extracted using water at the bligo and water ratio of 75%:25% (v/v). The slurry was filtered to obtain bligo fruit juice, which was then cooked at 100 °C for 10 minutes and added with 9% sucrose (w/v) and BPE at 1%, 2%, and 3%. Bligo juice without BPE addition was also prepared as a control.

2.3. Analysis

2.3.1. Water Content (Distillation)

Flasks were boiled and dried at 105 °C for 1 hour, followed by the addition of 3 ml of bligo juice and 75 ml of benzene solvent. The mixture was heated using an electric heater, refluxed slowly at low temperature for 45 minutes, and maintained in a hot state for 1 hour (AOAC, 2005). Moisture content was calculated using equation1:

$$\%\text{moisture content} = \frac{B-C}{B-A} \times 100\% \qquad (1)$$

where:
A = Weight of cup
B = Weight of cup and sample before drying (g)
C = Weight of cup and sample after drying (g)

2.3.2. Protein Content

Protein content was measured using Kjehdahl distillation method, and nitrogen value was converted to protein value using conversion factors (AOAC, 2005). The sample with as much as 0.5 g of wet material was placed in a Kjeldahl flask (50 ml capacity), added with 2 ml of sulfuric acid (93% + 98% free of N), heated in the fume hood until it became clear, and boiled for 30 minutes. After cooling, the wall of the Kjeldahl flask was washed with distilled water and boiled again for 30 minutes. After another cooling, the solution was added with 5–10 ml of distilled water and 6–15 ml of NaOH-$Na_2S_2O_3$ solution at 40:5 g and diluted with distilled water to 100 ml. Distillation was carried out using micro-Kjeldahl apparatus. The distillate was placed in an Erlenmeyer flask with 5 ml of 40% boric acid (saturated), which gave the indicator a methyl red mixture.

2.3.3. Fat Content

Approximately 1–2 g of the sample wrapped in filter paper and placed in a Soxhlet extraction tube attached to the Soxhlet distillate apparatus and the container flask with known weight. Cooling water was flowed through the condenser, and 15 ml of petroleum ether was added. Distillation was carried out for 4 hours. The flask containing fat extract was evaporated over a water heater and dried at 100 °C to constant weight. The residual weight in the bottle was weighed and expressed as the weight of fat and oil (AOAC 2005).

2.3.4. Ash Content

Ash content was determined according to AOAC 2005. A porcelain crucible was heated in muffle furnace, moved in desiccator, and weighed. Approximately 2 g of sample was heated on an electric stove until it became charcoal, incandescent in a muffle until it became a whitish ash, and finally weighed.

2.3.5. Carbohydrate Content

Carbohydrate content was measured indirectly by subtracting the weight of other constituents in the food including protein, fat, water, and ash from the total weight of food (AOAC, 2005). This value was referred to as total carbohydrate by difference and calculated using equation (2):

$$\text{total carbohydrate} = 100 - (\text{protein} + \text{fat} + \text{water} + \text{ash} + \text{ash} + \text{dietary fibre}) \qquad (2)$$

2.3.6. Vitamin C

Approximately 200–300 g of the sample was crushed in a waring blender until a slurry was obtained. The slurry was weighed, added with distilled water to a volume of 100 ml, and centrifuged to separate the filtrate. Afterward, 5 ml of filtrate was added with 2 ml of 1% starch solution and 20 ml of distilled water and then titrated using 0.01 N standard iodine (1 ml 0.01 N iodine = 0.88 mg ascorbic acid) (15).

2.3.7. Antioxidant Activity

Approximately 0.2 g of sample was added with 5 ml of methanol and then shaken vigorously using the vortex. Afterward, 0.2 ml of the extract was obtained, added with 2.8 ml of 0.1 mM DPPH reagent, and incubated in a dark room for 1 hour. Blank was prepared with 0.2 ml of methanol and 2.8 ml of 0.1 nM DPPH reagent. Absorbance was measured using a spectrophotometer at 515 nm wavelength (16).

2.3.8. Viscosity

The viscosity of the spreadable gel product was measured using a viscometer (Brookfield model DV1MLVTJ10, USA). Samples were measured using spindle 5 at 60 rpm and room temperature. The reading equilibrium time was 30 seconds (17).

2.3.9. Evaluation Sensory

The level of acceptance was determined using a 5-point hedonic test. Twenty-five panelists were asked to rate the color, flavor, viscosity, taste, and liking. The preference scale was 1 (very dislike), 2 (dislike), 3 (neutral), 4 (like), and 5 (like) (like very much).

3. Results and Discussion

3.1. Proximate Analysis

Proximate analysis of the water content, ash, protein, fat, and carbohydrate of bligo juice is shown in Table 1. The water content of bligo juice varied from 96.12% to 97.06% (Table 1). According to the FDA (18), any food or drink with a high water content typically has a moisture value of 85% or higher. This water content is normal and within the standard. The highest water content was obtained for the bligo juice with 3% BPE, followed by the control. Food products' shelf life is impacted by water migration and the accompanying change in moisture content through unfavorable changes to their physical, sensory, and microbiological properties (19). The moisture content of a product determines its shelf life and the viability of microorganism growth (20). However, further processing the juice through pasteurization, canning, or bottling can help eliminate microorganisms and increase its shelf life.

The protein content of bligo juices with 1% and 3% BPE was substantially higher than that of control. However, the bligo juice with 3% BPE had a protein content of 0.61%, which was smaller than the 0.80% and 0.85% of bligo juices with 1% and 2% BPE, respectively. In general, fruit juices contain a small amount of protein (21). Nevertheless, many plant compounds interact with proteins. According to Mazumder *et al*. (22), glutamic acid, aspartic acid, phenylalanine, leucine, threonine, serine, glycine, alanine, and valine can be found in the juice extracted from matured bligo fruit.

The ash content of bligo juice with BPE addition was in the range of 0.62%–0.41%. Total ash content was within the expected range of 0.3%–2% for fresh fruit and vegetables (23). This finding implied that the bligo juice was in line with the standard. This ash content was relatively lower than that for blended orange juice (23) and blended pineapple and orange juice (24). Compared with the control, bligo juice with 2% BPE had significantly increased ash content. Ani and Abel (25) reported that the peel extract of *Citrus maxima* fruit has higher ash content than its juice. High concentrations of different mineral elements are anticipated in samples with high ash contents. The fat content of bligo juice is between 0.11% and 0.13%. Similar to its protein content, bligo juice has a small amount of fat. Fruits have a large proportion of water at 85% average but contains low fat, hence their suggested role as a component of diets to lose weight (26).

BPE addition significantly and dose-dependently increased the carbohydrate content of bligo juice from 1.93% in control to 2.09%, 2.27%, and 3.26% upon the addition of 1%, 2%, and 3% BPE, respectively. Fruit peels possess a relatively high content of pectin. Pectin is a complex polysaccharide consisting mainly of galacturonic acid units linked by α-(1$\rightarrow$4) linkages. Its content in apple pomace, citrus peel, mango peel, and banana peel ranges from 2.8% to 24.5% (27). Although not measured in this study, BPE may contain an appreciable quantity of pectin, which might have increased the carbohydrate content in bligo juice.

The large molecules of carbohydrates interact with water and form gels or thickened dispersion that affect viscosity. Owing to this ability, carbohydrate-based compounds (such as pectin, starches, and vegetable gums) have been widely used in the food industry as thickening and gelling agents (28). The results showed that BPE addition dose-dependently increased the viscosity of bligo juice (Table 1).

Carbohydrate-based constituents, such as dietary fiber, also have various health benefits. Dietary fiber intake reduces the risk of developing coronary heart diseases (29), stroke (30), hypertension(31), diabetes (32), cardiovascular (33), and certain gastrointestinal disorder (34). Furthermore, increasing the dietary fiber intake ameliorates serum lipid concentration (35), assists in weight loss (36), and enhances immune function (37). Therefore, the increased carbohydrate content of bligo juice provides a high content of carbohydrate-based constituents such as dietary fiber and pectin.

Table 1. Characteristics of bligo juice with BPE addition.

Indicator	Control	Bligo peel extract (BPE)		
		1%	2%	3%
Water content (%)	96.79 ± 0.00[c]	96.68 ± 0.03[b]	96.12 ± 0.03[a]	97.06 ± 0.03[d]
Ash content (%)	0.51 ± 0.37[b]	0.41 ± 0.01[a]	0.62 ± 0.01[c]	0.45 ± 0.03[a]
Fat (%)	0.14 ± 0.00[a]	0.11 ± 0.00[a]	0.13 ± 0.00[a]	0.11 ± 0.00[a]
Protein (%)	0.62 ± 0.00[a]	0.80 ± 0.02[b]	0,85 ± 0.00[c]	0.61 ± 0.02[a]
Carbohydrate (%)	1.93 ± 0.00[a]	2.09 ± 0.04[b]	2.27 ± 0.01[c]	3.27 ± 0.01[d]

Different letters in the same row indicated a significant difference.
One-way ANOVA, post hoc Duncan $p > 0.05$. Each value is the average of thee replicates.

3.2. Vitamin C

Vitamin C, also called L-ascorbic acid (L-threo-hex-2-enono-1,4-lactone, ascorbate), is an essential antioxidant molecule in plant, animal, and human metabolism. Although plants and many animals can synthesize ascorbate in the liver or kidney, humans have lost this ability (38–40). Fruits and vegetables are the major source of vitamin C; thus, consuming these foods may have an important impact on human nutrition. A generous intake of ascorbate has various health advantages, such as enhancing the immune system (41,42) and reducing the risk of cancer by increasing TET's activity (43), inducing oxidative stress in cancer cells, or improving the activity of various chemical treatments (44).

A study on whole and peeled apples showed that peeling decreased the total phenolic content, ascorbic acid content, and antioxidant capacity by 26%, 48%, and 18%, respectively (45). This finding suggested that most antioxidant compounds are present in the peel. Here, BPE addition at 1%, 2%, and 3% significantly increased the vitamin C content of bligo juice from 1.40 mg/100 ml to 1.68, 2.55, and 2.10 mg/100 ml, respectively (Table 2). We hypothesized that bligo peel may also contain an appreciable quantity of vitamin C; thus, BPE addition may improve the health benefits of bligo juice. The biological functions of bligo juice must be further explored.

3.3. Antioxidant Activity

Fruit and vegetable antioxidants may help protect cells against the oxidative stress caused by free radicals and reduce the risk of degenerative diseases, such as cardiovascular diseases and cancers (46,47). Phenolic compounds contribute to antioxidant activity due to their ability as hydrogen donors, reducing agents, singlet oxygen quenchers, and metal chelators. Owing to this ability and their health benefits, antioxidant compounds from various fruit and vegetable sources have been explored. Karadeniz *et al*. (2004) reported that the antioxidant activity ranges from 13.7% (pear) to 62.7% (pomegranate) in fruits and from 12.5% (onion) to 40.8% (red cabbage) in vegetables.

Our previous study reported that bligo peel contains various phenolic compounds with antioxidant activity (12). In accordance with this finding, the present work found a significant increase in antioxidant activity upon BPE addition as shown in Table 2. The antioxidant activity increased by two, three, and fourfold for 1%, 2%, and 3% BPE additions, respectively, compared with that of the control. This activity is relatively higher than that in grapes (26.6%), apple (25.7%), pear (13.7%), spring onion (15.7%), potato (14.2%), and onion (12.5%) (48). Therefore, BPE addition can improve the antioxidant activity of bligo juice and consequently increase its health benefits.

Table 2. Viscosity, vitamin C, and antioxidant levels of bligo juice with BPE addition.

Indicator	Control	Bligo peel extract (BPE)		
		1%	2%	3%
Viscosity (cP)	3.40 ± 0.00[a]	3.80 ± 0.00[b]	3.88 ± 0.04[c]	4.00 ± 0.00[d]
Vitamin C (mg/100 ml)	1.40 ± 0.00[a]	1.68 ± 0.00[a]	2.55 ± 0.21[c]	2.10 ± 0.00[b]
Antioxidant activity (%)	10.55 ± 0.11[a]	24.31 ± 0.06[b]	32.16 ± 0.05[c]	41.37 ± 0.11[d]

Different letters in the same row indicated a significant difference.
One-way ANOVA, post hoc Duncan $p > 0.05$. Each value is the average of three replicates.

3.4. Sensory Properties

Sensory properties are important for consumer preference and acceptability of new products (46, 47). Therefore, the sensory parameters including color, flavor, taste, viscosity, and overall acceptability of bligo juice were evaluated. The acceptance level of bligo juice after BPE addition is shown in Table 3. In general, BPE addition significantly increased the acceptance level of color and viscosity. No significant difference in flavor was observed. The higher the BPE addition, the lower the acceptance level of taste. According to the overall acceptance, the bligo juice with 1% BPE showed the highest acceptance level that is similar to that of the control.

An increase in color acceptance was strongly correlated with the amount of BPE addition. The bligo juice with 3% BPE had a dark brown color, and others were pale brown (Figure 1). In addition, Maillard reaction might have occurred during the pasteurization of bligo juice, resulting in a dark color. This reaction is caused by the interaction between reducing sugars and primary amine groups at high temperatures (49). This finding was in agreement with Sun *et al.* (1), who stated that bligo juices processed without peel are white or light gray in color.

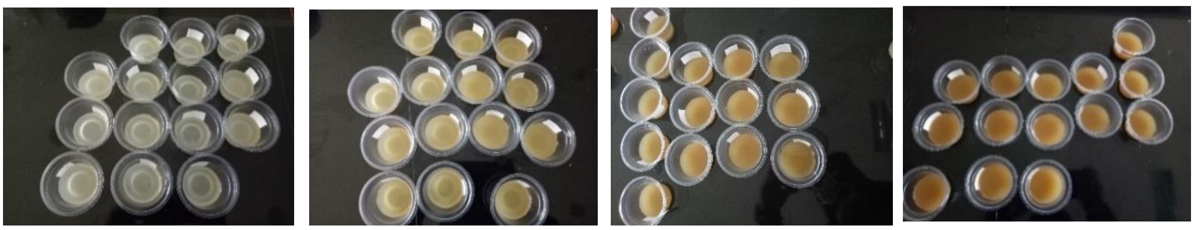
Table 3. Sensory profile of bligo juice with BPE addition.

Indicator	Control	Bligo peel extract (BPE)		
		1%	2%	3%
Color	1.20 ± 0.41^{a}	2.32 ± 0.47^{b}	3.32 ± 0.55^{c}	4.04 ± 0.72^{d}
Flavor	3.08 ± 1.13^{a}	3.04 ± 0.87^{a}	3.32 ± 1.12^{a}	3.12 ± 1.39^{a}
Taste	4.04 ± 0.82^{b}	3.76 ± 0.71^{b}	3.64 ± 0.97^{b}	3.04 ± 1.18^{a}
Viscosity	1.80 ± 0.89^{a}	2.44 ± 0.57^{b}	2.96 ± 0.66^{c}	3.16 ± 0.83^{c}
Acceptability	3.16 ± 0.86^{c}	2.68 ± 0.86^{bc}	2.28 ± 0.76^{ab}	1.84 ± 1.03^{a}

The different letters in the same row indicated a significant difference.
One-way ANOVA, post hoc Duncan $p > 0.05$. Each value is the average of 25 panelists.

In terms of flavor, the panelist preferred the bligo juice with 3% BPE, followed by the control. Flavor compounds form as secondary metabolites of fatty or amino acid precursors (carbonyl compounds, carboxylic acids, alcohols, and lactones) and carotenoids or during fruit ripening (terpenes esters, and ethers) (50). According to Sun *et al*. (1), the "grassy" and "cooked-vegetables" flavor of bligo juice is possibly due to the high content of volatile compounds such as 1,4-butanediol, 5-methyl-2-hexanone, 2-methylfuran, 2-ethylfuran, 2-pentylfuran pentanal, (E)-2-hexenal, pentanol, (Z)-2-hexenol, heptanal, octanal, and hexanal from the peel.

The higher the BPE addition, the lower the acceptance level of taste. The peel contributes to acidity, bitterness, and astringency (51). According to Vermeulen *et al*., the high concentration of sulfur volatile compounds (which is typically found in many other vegetables, particularly cucurbits) increases the risk of an unpleasant taste in juice. Bhardwaj and Pandey (52) suggested mixing two or more fruit and vegetable juices to create a ready-to-serve beverage as a handy substitute for eating foods with high acidity, astringency, or bitterness (52).



Control BPE 1% BPE 2% BPE3%

Figure 1. Color of bligo juice with BPE addition.

In terms of viscosity, the bligo juice with BPE addition tended to be preferred by the panelists. A study on the sugar composition of pectin in bligo juice discovered that interactions between acidic polysaccharides and galactans could increase its viscosity (53). Overall, the panelists preferred the bligo juice with 1% BPE.

4. Conclusions

BPE addition increased the vitamin C levels, antioxidant activity, carbohydrates, fats, and acceptance of fruit juice color of bligo juice. The addition of 1% BPE had the highest acceptance rate among the variations. On the basis of the results, BPE addition increased the vitamin C, antioxidant activity, and carbohydrate content of bligo juice, thus improving its health benefits.

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Author Contributions

D.N.H. conceptualization, methodology, investigation, writing-original draft, data curation; A.F. supervision, data curation; S.W. writing original draft; T.S. methodology investigation; N.H. supervision method, writing -original draft.

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Conflicts of Interest

The authors declare no conflict of interest.

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