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Characteristics of Yogurt Ice Cream Fortified with Red Dragon Fruit Puree as Anti-Obesity Functional Food

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ABSTRACT

This study aimed to determine the effect of Red Dragon Fruit Puree (RDP) added to yogurt ice cream on its sensory, physical and chemical characteristics as well as its anti-obesity effect on mice fed with high-fat diet. To obtain the optimal addition level, RDP was added to the yogurt-based ice cream dough in various proportions: 0 (control), 10, 20, 30, and 40%. The sensory evaluation signifies that 30% RDP addition results in the best yogurt ice cream because it produces favorable flavor, attractive color (purplish pink), and the most preferable taste for the panelist. Data on physical and chemical characteristics showed that RDP yogurt ice cream has lower overrun, total solid, fat, carbohydrate, and calorie content than non-RDP yogurt ice cream (p<0.05). The animal test identified that consumption of RDP yogurt ice cream for 14 days could reduce the mice's (a xenograft model) body weight under normal and high-fat diet conditions (p<0.05). Thus, RDP yogurt ice cream with specific physical and chemical characteristics potentially results in anti-obesity effects in pre-clinic setting. RDP yogurt ice cream merits further studies, especially for clinical test, before the commercialization stage.

Keywords: antiobesity, ice cream, red dragon fruit, yogurt

INTRODUCTION

Ice cream is a dairy product greatly demanded by all walks of life. However, ice cream on the market contains high fat and sugar; therefore, people with obesity or on a diet program should avoid consuming ice cream (Su'i et al. 2020). Nowadays, yogurt ice cream has become a trend because in addition to its taste, yogurt ice cream also offers health benefits. Yogurt is a fermented dairy product using Lactic Acid Bacteria (LAB), namely, Lactobacillus bulgaricus and Streptococcus thermophillus (Aktar 2022). Yogurt is rich in protein, vitamins, minerals, and good bacteria beneficial for health. In contrast to other dairies, people with lactose intolerant can also consume yogurt because the fermentation process changes lactose to lactic acid, making it suitable for them (Yanni et al. 2020). Previous studies have stated that yogurt efficaciously reduces the risk of several significant chronic diseases, such as heart disease, diabetes, and cancer, and is healthy for consumption for obese people (Baspinar & Güldaş 2021). However, yogurt has a plain taste, less attractive appearance, and overpowering aroma of milk. Thus, some people do not like yogurt's plain taste.

Yogurt commonly consumed by adding pieces of fruit to improve the taste and add health benefits on the gut's health (Fernandez & Marrete 2017). Dragon fruit is a fruit with delicious taste and an attractive color. Besides its attractive appearance and sweet taste, dragon fruit has a high fiber content. Dragon fruit ontains betacyanin and many other bioactive ompounds, such as vitamins C, B, and E, flavonoids, carotenoids, and polyphenols, which are potential antioxidants (Fatmawati et al. 2018). Another advantage of red dragon fruit is rich in fiber, which is beneficial for people with obesity and diabetes. One hundred grams of dragon fruit contains 10.1 g of fiber, which can reduce fat absorption in the body (Fadlilah et al. 2021). Song et al. (2016) found that the betacyanin compound in dragon fruit significantly reduced weight gain in rats fed with a high-fat diet and 200 mg/kg of red dragon fruit for 14 days. However, there has been no research applying dragon fruit to food products, such as yogurt, which can function as

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an anti-obesity food. Therefore, this study aimed to determine the effect of adding Red Dragon Fruit Puree (RDP) with different concentrations on physical characteristics, nutrient content, and sensory characteristics of the yogurt ice cream. Further, this study also explored the effect of RDP in yogurt, as a functional food, on body weight of mice as the obesity test animal model

METHODS

Design, location, and time

The study consisted of four stages. The first was the organoleptic test, followed with physical and chemical characteristics tests and the final stage was in vivo test using mice models. The in vivo test for the anti-obesity trial employed a Completely Randomized Design (CRD). This study employed five treatments (T0: control, T1-4: 10, 20, 30, and 40% addition of red dragon fruit) and four replications. The best treatments according to the organoleptic test then was tested on mice for the in vivo test. The mice models were assigned randomly into the two groups: treatment group and control group.

This research was carried out at the Food Chemistry and Nutrition Laboratory, Faculty of Animal Husbandry and Agriculture, Universitas Diponegoro in September-December 2019. The study protocol was approved by the Faculty of Medicine, Universitas Diponegoro, using a method based on the Ethical Clearance of No.127/ EC/H/KEPK/FK-UNDIP/X//2019.

Materials and Tools

The main ingredients of this study were cow's milk, yogurt starter (*biokul*), red dragon fruit, sugar, and skimmed milk. The tools used in this research were a container, filter cloth, blender, analytical balance (DJ Excellent Scale, USA), stopwatch, oven, desiccator, Bunsen, porcelain cup.

Procedures

Preparation of red dragon fruit puree (RDP)

Red dragon fruit puree was made by cutting the red dragon fruit into four parts and taking the flesh by separating it from the skin and chopping it into pieces. The red dragon fruit was weighed according to the treatment and was then blended until became pulp. Then, it was filtered using a filter cloth. The formulations of RDP yogurt ice cream were 500 ml of milk, 15 g of yogurt starter, 25 g of sugar, 30 ml of skim milk, and red dragon with each treatment of T0-4: 0; 50; 100; 150; and 200 g.

Preparation of red dragon fruit yogurt ice cream (RDYIC)

To make the yogurt, 500 ml fresh cow's milk was pasteurized for 15 minutes at 72°C. Then, it was put into a sterilized container or jar. Afterward, the dragon fruit puree was mixed according to the treatment T1-4: 10, 20, 30, and 40% of the addition of red dragon fruit from the total milk used (v/v). The mixture was then stirred until homogeneous. The bacterial starter was weighed for 3% of 500 ml of milk and then inoculated into a jar containing pasteurized milk and fresh dragon fruit puree. The jars were tightly closed and incubated for 12 h at 43°C. The next step was adding sugar and skim milk. Then, the mixture was stirred well, mixed with the yogurt mixture, and finally pasteurized again for 30 minutes at a temperature of 85°C. The produced yogurt ice cream was then put into a freezer at -18°C for approximately 15 h prior to further analysis.

Sensory test

Organoleptic testing was carried out using an objective sensory measurement with the attribute rating test of taste, color, texture, and aroma; in addition it also included the overall acceptance. The assessment was carried out using 25 semi-trained panelists. The taste attribute employed five levels: 1 (very sour), 2 (sour), 3 (quite sour), 4 (slightly sour), and 5 (not sour). Yogurt aroma attributes used five levels: 1 (not fruity), 2 (slightly fruity), 3 (fairly fruty), 4 (fruity), and 5 (very fruity). The color attribute employed five levels: 1 (not purple), 2 (slightly purple), 3 (quite purple), 4 (purple), and 5 (strong purple). Texture attributes have five levels: 1 (not soft), 2 (slightly soft), 3 (quite soft), 4 (soft), and 5 (very soft). Meanwhile, the overall acceptance of the preference used a scale of 1 (dislike), 2 (like slightly), 3 (like moderately), 4 (like), and 5 (likes extremely). The best RDYIC formula was measured by the highest overall organoleptic score. The RDYIC which scored best in the sensory test was then tested for physical and chemical analysis as well as in-vivo test on mice.

Physical characteristics test

The physical characteristic test of ice cream included the overrun analysis (Goff & Hartel 2013) and the total solid analysis (Singo & Beswa 2019). The overrun measurement was done by measuring the ice cream dough with the same volume using a measuring cup before and after the process with an ice cream maker. Total solids analysis was conducted by calculating the moisture content. Afterward, the moisture content was used to calculate the total solids by deducting 100% with the percentage of moisture content.

Nutrient content analysis

The test for chemical characteristics of ice cream included proximate and calorie content tests. The proximate tests measured the moisture content and ash content following the AOAC method (Wong *et al.* 2019), the protein content based the Kjeldahl method (Chen *et al.* 2019), the fat content following the Soxhlet method, and the carbohydrate test by difference (Góral *et al.* 2018). The calorie content test employed the proximate test results by calculating the number of carbohydrates, fats, and proteins with 9 kcal of calories produced by 1 g of fat and 4 kcal calories produced by 1 g of protein or 1 g of carbohydrates (Balthazar *et al.* 2017).

Pre-clinical test of red dragon fruit yogurt ice cream as anti-obesity functional food

The pre-clinical test (in vivo) was done on mice as experimental animals and was conducted to determine the effect of RDPYIC with the best organoleptic test result on the mice's weight gain. The pre-clinical test has been approved by the Ethical Clearance No.127/EC/H/KEPK/ FK-UNDIP/X//2019. The pre-clinical trial was conducted in 12 mice aged 2-3 months and weighed 20-35 g. These mice were obtained from mouse breeders in Banyumanik. Pre-clinical trials were conducted for five weeks using a force-feeding method. The variety of feed given was Normal Feed (ND) and High-Fat Feed (HFD). The ND group received commercial feed while the HFD group received a mixture of commercial feed and high-fat components: egg yolk and coconut oil. The provision of yogurt ice cream was adjusted to an acceptable dose or the Acceptable Daily Intake (ADI) converted according to the mice's weight with 0.728 g/weight of mice. The normal diet treatment referred to providing commercial feed to the mice while the high-fat diet referred to providing a mixture of commercial feed (80%), egg yolk (10%), and coconut oil (10%).

The pre-clinical test was conducted using the optimal treatment based on the organoleptic test, which was additon of 30% of red dragon fruit. The rest of the feed was weighed every day, and the body weight of the mice was measured at the beginning and end of the treatment.

Data analysis

The experimental method of this research was a Completely Randomized Design (CRD) with a single factor of five treatments and four replications. The physical and chemical characteristics data (overrun, total solids, proximate, and calorie content) were compared between the best score in RDYIC formula and regular yogurt without any RD addition using t-test. The organoleptic test data were analyzed using the Kruskal Wallis non-parametric test. If an effect of treatment on the organoleptic properties of yogurt ice cream had been found, a Mann-Whitney follow-up test was carried out. The data for pre-clinical trials were statistically analyzed using the t-test. All obtained data were analyzed using the SPSS 22.0 at a significant level of 0.05.

RESULTS AND DISCUSSION

Sensory of characteristics red dragon fruit puree yogurt ice cream

The results of the organoleptic test of red dragon fruit puree yogurt ice cream are presented in Table 1. The RDP addition affects the aroma, taste, color, texture, and the overall acceptance of yogurt ice cream. The dominant aroma and taste of yogurt in the treatment T0 (control) resulted from the main ingredient, namely fermented cow milk. Therefore, the yogurt has a characteristic taste and sour aroma that are produced during the milk fermentation process.

Table 1 shows that non-RDP yogurt higher rates in term texture and aroma compared to all RDP treatments and the difference was statistically significant. The 20% RDP had the highest rate in taste but it was not statically different compared to other RDP treatments. The 10% formula had the highest score in texture but it was not statistically different compared to other RD treatments. The 40% formula had the highest rate in color compared to other formulas and the

Table 1. Sensory characteristics of red dragon fruit puree yogurt ice cream

Attributes	Addition of red dragon puree (%)				Rating	
	0	10	20	30	40	interpretation (1-5)
Aroma	$4.32{\pm}0.46^{\rm a}$	$3.12{\pm}0.46^{\text{b}}$	$2.82{\pm}0.51^{\text{b}}$	$3.10{\pm}0.72^{\text{b}}$	$2.31{\pm}0.15^{\text{b}}$	Not fruity-Fruity
Taste	2.12±0.82ª	$3.10{\pm}0.55^{\text{b}}$	$3.11{\pm}0.51^{b}$	$3.07{\pm}0.32^{\rm b}$	$3.09{\pm}0.53^{\text{b}}$	Very sour-Sour
Color	$1.58{\pm}0.59^{\mathrm{a}}$	$2.76{\pm}0.23^{\text{b}}$	$3.22{\pm}0.48^{\rm c}$	$3.72{\pm}0.41^{\text{d}}$	$4.46{\pm}0.48^{\circ}$	Not purple-Strong purple
Texture	$4.10{\pm}0.55^{a}$	$3.29{\pm}0.44^{\text{b}}$	$3.13{\pm}0.57^{\text{b}}$	$3.54{\pm}0.69^{\text{b}}$	$3.58{\pm}0.25^{\text{b}}$	Not soft-Very soft
Overall acceptance	3.51±0.47 ^{abc}	3.22±0.65°	3.28±0.72 ^{bc}	4.32±0.78ª	3.82±0.69 ^{ab}	Dislike–Like extremely

The data were expressed as mean±standard deviation

Different superscripts in the same row show a significant difference (p<0.05)

differences statistically significant. The more RDP added to the yogurt ice cream, the stronger the aroma and color. Thus, RDP affects the taste, aroma, color, and texture differently. Hence, the overall score for all 5 formulas showed that the T3 RDP treatments with 30% addition of RDP received the highest score and it was most preferred in terms of overall acceptance and it was used for further test.

The sweet taste and the distinctive aroma of red dragon fruit may disguise the aroma of yogurt. However, red dragon fruit has a strong unpleasant aroma; thus, the addition large ammount of dragon fruit juice could reduce the panelists' acceptance. Meanwhile, the taste of yogurt can be influenced by the addition of other ingredients, such as honey and low-calorie sweetener functioning as a sweetener. Red dragon fruit has a fairly sweet taste; therefore, this fruit is suitably made into a yogurt ice cream fortifier that can compensate the sour taste in vogurt. The sour taste in yogurt occurs during the fermentation process by LAB, which produces lactic acid, citric acid, and acetic acid; thus, the lower the pH of fermented milk, the sourer the yogurt taste (Andila & Pato 2018). In the control treatment, the resulted taste was too sour, which affected the level of panelists' acceptance.

Higher addition of red dragon fruit puree also significantly affected the samples' color because the addition of RDP produce stronger color of purple. The stronger color was produced by the increasing betacyanin pigment content in the ice cream (Thaiudom *et al.* 2021). The addition of red dragon fruit puree produce a slightly softer yogurt ice cream texture because red dragon fruit has a high moisture content, which can form ice crystals during freezing.

Physical characteristics of yogurt ice cream with red dragon fruit addition

Table 2 shows that the overrun and total solids values of RDP yogurt ice creambetween T0 and T3 formula were significantly different (p<0.05). The overrun value of yogurt ice cream with the addition of RDP decreased significantly than the control. The decrease in overrun value was caused by red dragon fruit juice, which had a fairly high fiber content. The high fiber content in fruits results in decreased overrun value because fiber can affect the viscosity of the ice cream mix, which binds air; thus, ice cream mix is difficult to expand (Hapdang et al. 2021). In addition, the fiber in red dragon fruit can bind water so that the ice cream mix got thick and as a result, the total solids of the ice cream added with RDP would also decrease. Therefore, more quantity of red dragon fruit puree added would add more moisture into the ice cream content and reduced the total solids. Masykuri et al. (2012) stated that the lower the total solids, the greater the amount of frozen water; thus as a result, less air could enter and cause ice cream overrun to be low.

 Table 2. Physical characteristics of red dragon fruti puree yogurt ice cream

	Treatment		
Parameters	Yogurt ice cream	Red dragon fruit puree yogurt ice cream (30%)	
Overrun (%)*	21.57±2.35	14.32±1.37	
Total Solids $(\%)^*$	44.83±2.87	36.23±2.57	

*Significant differences between yogurt ice cream versus red dragon pure yogurt ice cream (p<0.05)

Nutrient content of red dragon fruit puree yogurt ice cream

The results of the nutrient analysis are presented in Table 3. The results of the independent sample t-test show that the addition of red dragon fruit puree had a significant effect on moisture, fat, carbohydrate, and calorie content but there was no significant effect on ash and protein content. The differences in the moisture content of the two formulas were caused by the addition of red dragon fruit puree, which had3 a fairly high moisture content. According to Herianto et al. (2015), red dragon fruit flesh had 85.77% of moisture content. The moisture content in ice cream tended to increase when the addition of red dragon fruit puree was increased. The ash content in both treatments had a fairly low value. The protein content of vogurt ice cream increased along with the addition of red dragon fruit puree because dragon fruit contained 0.53% protein (Nguyen et al. 2021). This result is in line with Nurul and Asmah (2014), who found 100 g of red dragon fruit flesh contained 0.159-0.229 g of protein.

The significance decrease in fat content of red dragon fruit puree yogurt ice cream happened because the RDP concentration added in the yogurt was quite large, namely 30%. Red dragon fruit has a very low-fat content of 0.6 g per 100 g; thus, greater RDP addition in yogurt will reduce the fat content of yogurt (Zahro & Nisa 2014). In addition, during the fermentation process, fats undergo hydrolysis into simpler compounds by lipase that are produced by lactic acid bacteria (Triasih & Priyadi 2021). The decreasing carbohydrate levels in red dragon fruit puree yogurt ice cream were influenced by different methods in yogurt ice cream. Lactic acid bacteria convert carbohydrates in the sample into simple sugars that are used as an energy source; thus, carbohydrates in the form of natural sugars would decrease. Julianto (2016), stated that lactic acid bacteria in yogurt convert carbohydrates into monosaccharides, which will be used to produce lactic acid. The total calorie content of sample with 30% red dragon fruit puree decreased because T3 had lower fat and carbohydrate content than T0.

Anti-obesity effect of red dragon yogurt ice cream

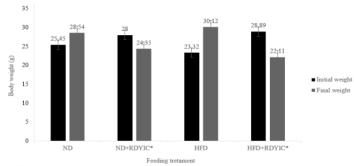
Preclinical test was carried out to examine the effect of RDP yogurt ice cream on changes of body weight of mice. The T3 or 30% addition of red dragon fruit puree into yogurt ice cream was given for the treatment group. All groups that were treated for 14 days showed a significant difference in initial weight and final weight, except for the mice treated with Normal Diet (ND) only. The administration of a normal diet for 14 days did not significantly change the weight of the mice, but the administration of RDP yogurt ice cream with ND significantly decreased the final weight of the mice (p<0.05). Meanwhile, the HFD treatment significantly increased the body weight of mice. On the other hand, feeding HFD combined with RDP yogurt ice cream showed significant reduction in the body weight of mice. Thus, the weight loss experienced by mice with ND and HFD diets could be associated with the consumption of RDP yogurt ice cream (Figure 1).

Adhi *et al.* (2018) have proven that fiber in red dragon fruit inhibits fat absorption and allows them to be eliminated with feces. Soluble fiber forms a thick solution that inhibits the digestion

Table 3. Chemical characteristics of red dragon fruit puree yogurt ice cream

	Treatment			
Parameters	Yogurt ice cream	Red dragon fruit puree yogurt ice cream (30%)		
Moisture content (%)*	52.11±1.85	64.70±0.78		
Ash Content (%)	$0.72{\pm}0.24$	$0.67{\pm}0.02$		
Protein Content (%)	$2.36{\pm}0.74$	3.12±0.13		
Fat Content (%)*	8.18±0.52	5.36±0.21		
Carbohydrate content (%)*	34.88±2.73	22.11±0.74		
Calorie content (kcal/g)*	222.58±8.44	149.16±4.51		

*Significant difference between yogurt ice cream and red dragon fruit puree yogurt ice cream (p<0.05)



The mice were fed with a Normal Diet (ND); Normal diet+30% Red Dragon Fruit Puree Yogurt Ice Cream (ND+RDYIC); High-Fat Diet (HFD) and; High-Fat Diet+30% Red Dragon Fruit Puree Yogurt Ice Cream (HFD+RDYIC) The data are expressed as a mean of ± standard deviation. *Significant difference between the initial weight and final weight

Figure 1. Comparison of mice's body weight before and after the feeding treatment for fourteen days

and absorption of carbohydrates and fats. Otherwise, fiber can be fermented by intestinal flora into Short-Chain Fatty Acids (SCFA); one of them is butyrate, the main SCFA in the human intestine. SCFA production will affect appetite either directly in the brain or via different signals (Astuti & Jenie 2020; Hjorth et al. 2019). Consuming fiber prolong the period of fullness due to the expansion of the large intestine. Dietary fiber also promotes growth of beneficial microbes in the gut, particularly lactobacilli. Gut microbiota can influence bile acid metabolism and produce various metabolites, including SCFA and neuroactive. Meanwhile, negligible protein sequences can be translocated to the peripheral circulation or interact with enteroendocrine cells and systems, which causes release of neuropeptides and peripheral hormones related to appetite and eating behavior (Esmaeili et al. 2022). Consumption of fiber regularly can help to lose weight while daily intake of yogurt can help reduce visceral fat by 81% since the high protein content in yogurt helps the body to metabolize fat faster (Michels et al. 2020).

CONCLUSION

Addition of RDP to yogurt ice cream produced red dragon fruit puree yogurt ice cream (RDYIC) with more fruity aroma, less sour taste, more purple color, and softer texture. Yogurt with 30% RDP was the most preferred compared to other formulas. RDYIC had a high moisture content with low overrun and total solids values, fat content, carbohydrate content, and total calorie content. Preclinical (in vivo) test found that the body weight of mice decreased when they consumed the RDYIC for 14 days in addition to both normal diet and high fat diet. Thus, RDYIC has a potential as functional food to be used as dessert product with an anti-obesity property.

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DECLARATION OF INTERESTS

The authors have no conflict of interest.

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