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The Effect of pandan leaf (*Pandanus Amaryllifolius*) at different concentrations on the physical properties of yogurt

Eka Wulandari, Andry Pratama
Department of Livestock Product Technology, Faculty of Animal Husbandry, Padjadjaran University, Jl Raya Bandung, Sumedang, Jatinangor, West Java, Indonesia
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Abstract
This study aimed to determine the effect of adding pandan leaf extract (*Pandanus Amaryllifolius*) at different concentrations on the physical properties of yogurt. A Completely Randomized Design (CRD) was employed with four treatments of pandan leaf extract concentrations: 0% (P0), 5% (P1), 10% (P2), and 15% (P3). The observed parameters included L*, a*, b* color values, water holding capacity, and syneresis. The data were analyzed using analysis of variance (ANOVA) followed by Duncan's multiple range test. The results showed that the addition of pandan leaf extract had a significant effect ($P<0.05$) on the color and water holding capacity of yogurt, but had no significant effect ($P>0.05$) on yogurt syneresis. Based on these findings, the addition of pandan leaf extract up to a concentration of 15% was able to improve the physical characteristics of yogurt, particularly in terms of color and water holding capacity.

Keywords: Yogurt, *Pandanus Amaryllifolius*, water holding capacity, syneresis, color

Introduction
Yogurt is a fermented milk product produced using lactic acid bacteria (Lactobacillus bulgaricus and Streptococcus thermophilus), with or without the addition of other food ingredients (BSN, 2009). Yogurt contains essential fatty acids, vitamin A, vitamin B2, and vitamin B12 which are beneficial to human health (Hadjimbei *et al*., 2022) [6]. Yogurt can be classified into two types, namely plain yogurt (without flavoring) and flavored yogurt with the addition of fruit pieces (Corrieu, 2016) [2]. Indonesia has a long-standing tradition of consuming plant-derived spices for their health benefits. These spices are commonly regarded as "herbal" remedies by the public and are believed to have fewer negative side effects compared to chemical-based medicines (Saleh *et al*., 2023) [12]. One of the spice plants widely found and commonly used by Indonesian people is pandan leaf (*Pandanus Amaryllifolius*) (Silalahi, 2018) [14]. Pandan leaves contain flavonoids, alkaloids, saponins, and antioxidants that are beneficial to human health (Dalimartha, 2008) [3]. The addition of pandan leaf (*Pandanus Amaryllifolius*) to yogurt is expected to improve its physical quality. Physical characteristics such as syneresis, water holding capacity, and color are among the important factors determining the quality of yogurt produced (Manab, 2008) [10].

Research Methods
Preparation of Pandan Leaf Extract
The preparation of pandan leaf extract was carried out by cutting the pandan leaves into small pieces, followed by blending with the addition of distilled water at a ratio of 1:1 until a smooth consistency was obtained. The blended pandan leaves were then filtered to separate the remaining leaf residues. The pandan leaf extract was subsequently heated at 80°C for 10 minutes to eliminate any bacteria still present in the extract..

Yogurt Preparation
Yogurt preparation was conducted based on the method of Starbard (2015) [15] with several modifications. Fresh cow's milk was supplemented with 5% (w/v) skim milk powder, followed by pasteurization at 80°C for 10 minutes. After pasteurization, the mixture was cooled to 40°C, and pandan leaf extract was added at concentrations of 0%, 5%, 10%, and 15% (v/v). A starter culture consisting of Lactobacillus bulgaricus and Streptococcus thermophilus was added at 3% (v/v) of the total yogurt mixture volume. The mixture was then incubated at 37°C for 18 hours until yogurt was formed.

Color Analysis
The color analysis of yogurt was conducted using a CS-10 Colorimeter following the method described by Zulaikhah and Fitria (2020) [17]. Yogurt samples were placed in transparent containers with a volume of 50 mL. Color measurements were carried out by attaching the CS-10 Colorimeter to the surface of the container. Data were collected from three different parts of the yogurt for each treatment. The L* value indicates the degree of brightness of the resulting color (L*:0 indicates black, while L*:100 indicates pure white). The a* value represents the degree of redness or greenness, whereas the b* value represents the degree of yellowness or blueness (Suliasih *et al*., 2016).

Water Holding Capacity Analysis
The water holding capacity analysis of yogurt was performed using the centrifugation method according to Hassan *et al*. (1996) [8]. A total of 2 mL of yogurt sample was placed into a centrifuge tube and centrifuged at 4000 rpm for 10 minutes. The resulting supernatant was carefully separated by slowly pouring it off until it was completely separated from the yogurt precipitate. The yogurt precipitate was then weighed. The water holding capacity was calculated based on the difference between the initial

sample weight and the supernatant obtained using the following formula:

$$WHC\ \% = \frac{(Initial\ Sampel\ -\ Supernatan)}{Initial\ Sampel}\ X\ 100$$

Syneresis analysis

Syneresis analysis was conducted using the drainage method to measure spontaneous syneresis without external influence by placing 25 mL of yogurt sample on No. 40 filter paper for 30 minutes at room temperature. The separated whey was then measured using a measuring cylinder (Lee and Lucey, 2010) [9]. The percentage of syneresis was calculated based on the difference between the initial sample weight and the whey obtained using the following formula:

$$Sineresis\ \frac{(Initial\ Sample\ -\ whey)}{Initial\ Sample}\ X\ 100$$

Statistical Analysis

This study was conducted using a Completely Randomized Design (CRD) consisting of four treatments of pandan leaf extract addition, namely 0%, 5%, 10%, and 15%. The data obtained were analyzed using analysis of variance (ANOVA). Further analysis was carried out using Duncan's Multiple Range Test (Gasperz, 1995) [5].

Hasil & Pembahasan

1. L* Color Analysis

The results of the color analysis of yogurt with the addition of pandan leaves (*Pandanus Amaryllifolius*) at different concentrations on brightness (L*), redness or greenness (a*), and yellowness or blueness (b*) are presented in Table 1.

Table 1: Effect of Pandan Leaf Extract Treatment on Yogurt Color

Treatment	Color		
	L*	a*	b*
P0	83.14[a]	-3.25[a]	6.69[a]
P1	77.92[b]	-5.28[b]	10.39[b]
P2	76.33[c]	-5,91[c]	12.63[c]
P3	73,59[d]	-6.56[d]	13.87[d]

Based on Table 1, the addition of pandan leaf extract to yogurt had a significant effect on the brightness of the yogurt color produced. The addition of pandan leaf extract tended to decrease the L* brightness value of yogurt as the concentration of pandan leaf extract increased. This change in yogurt brightness may be attributed to the chlorophyll content of pandan leaves dissolved in the yogurt during the fermentation process (Hörtensteiner and Kräutler, 2011) [7]. Chlorophyll contains derivative compounds in the form of pheophytin, which produces a dark brownish color, thereby reducing the brightness level of yogurt. Pheophytin may be formed due to heat exposure, acidic conditions, enzymatic changes, and the fermentation process (Dimara *et al*., 2012).

2. a* Color Analysis

Based on Table 1, the addition of pandan leaf extract to yogurt had a significant effect on the red-green color component (a*) produced. Lower a* values indicate a greener color, whereas higher a* values indicate a redder color. According to the data presented in Table 1, the green color intensity increased as the concentration of pandan leaf extract increased. This may be attributed to the chlorophyll content in pandan leaves, which is responsible for providing the green coloration and increased along with the higher concentration of pandan leaf extract added (Hörtensteiner and Kräutler, 2011) [7].

3. b* Color Analysis

Based on Table 1, the addition of pandan leaf extract to yogurt had a significant effect on the blue-yellow color component (b*) produced. Lower b* values indicate a bluer color, whereas higher b* values indicate a yellower color. According to the data in Table 1, the yellowness of the yogurt tended to increase. This may be caused by the increased chloroplast pigments a and b in chlorophyll, which function as coloring agents in pandan leaves (Puspita *et al*., 2021) [11].

4. Water Holding Capacity Analysis

The results of the water holding capacity analysis of yogurt with the addition of pandan leaf extract (*Pandanus Amaryllifolius*) at different concentrations are presented in Table 2.

Table 2: Effect of Pandan Leaf Extract Treatment on the Water Holding Capacity of Yogurt

Treatment	Water Holding Capacity (%)
P0	19.79[b]
P1	19.99[b]
P2	22.11[a]
P3	21.99[a]

Based on Table 2, the addition of pandan leaf extract to yogurt had a significant effect on the water holding capacity of the yogurt produced. The addition of pandan leaf extract tended to increase the water holding capacity of yogurt. The increase in water holding capacity may be attributed to the cellulose content of pandan leaves, which contains hydroxyl groups (-OH) capable of forming hydrogen bonds with water molecules (Sheltami *et al*., 2012) [13]. The hydroxyl groups present in the cellulose of pandan leaves can improve the water holding capacity of yogurt due to their hydrophilic properties (Diyana *et al*., 2021) [4]. The presence of fiber in pandan leaves (*Pandanus Amaryllifolius*) added to yogurt may also assist the milk coagulation process during fermentation, thereby allowing more water to bind to the protein structure of milk.

5. Syneresis Analysis

The results of the syneresis analysis of yogurt with the addition of pandan leaf extract (*Pandanus Amaryllifolius*) at different concentrations are presented in Table 3.

Table 3: Effect of Pandan Leaf Extract Treatment on Yogurt Syneresis

Treatment	Syneresis (%)
P0	28,25[a]
P1	31.25 [a]
P2	32.25 [a]
P3	33.50 [a]

Based on Table 3, the addition of pandan leaf extract to yogurt did not have a significant effect on the syneresis value of the yogurt produced. The addition of pandan leaf extract tended to slightly increase yogurt syneresis;

however, the effect was not statistically significant. The increase in syneresis value may be caused by the increased water content in yogurt as the concentration of pandan leaf extract increased. Higher water content in yogurt may result in the hydroxyl groups in pandan leaf extract being unable to bind all of the water present, thereby slightly increasing the syneresis value of yogurt (Diyana *et al*., 2021) [4].

Conclusion

The addition of pandan leaf extract (*Pandanus Amaryllifolius*) at different concentrations in yogurt had a significant effect on the color and water holding capacity of the yogurt, but did not significantly affect the syneresis of the resulting yogurt. The addition of higher concentrations of pandan leaf extract tended to decrease the brightness level (L*) while increasing the intensity of green (a*) and yellow (b*) colors in yogurt. The addition of pandan leaf extract was able to improve the water holding capacity of yogurt due to the presence of hydroxyl groups (-OH) and cellulose compounds in pandan leaves, which are hydrophilic and capable of binding water in yogurt. However, the addition of pandan leaf extract did not have a significant effect on yogurt syneresis.

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