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Evaluation of physicochemical properties and fatty acid profile on Indonesian local beef tallow and lard using two different rendering methods

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Abstract

Tallow and lard are rendered products that are useful for nutritional, feed, and pharmaceutical use due to their high content of essential fatty acids. However, the quality of Indonesia's local beef tallow and lard does not exist in terms of the basic information and effectiveness of these products. This study therefore aimed to evaluate and present information on the rendered fats obtained from two different rendering methods of local beef tallow and lard on physicochemical properties and fatty acid profiles. The yield, physicochemical properties, and fatty acid profile of beef tallow and lard rendered via dry (DR) and wet (WR) rendering at 90-100°C for 30 mins were evaluated. The dry- and wet-rendering methods exhibited the highest yields. WR methods of both samples have a brighter yellow color than DR methods. For physicochemical properties, the WR method had the highest acid value ($P<0.05$), whereas the saponification value and peroxide value were not significantly different for both rendering methods. There were significant differences in fatty acid compositions for tallow and lard prepared from the two rendering methods. Based on the yield and physicochemical properties of the tallow, both methods fulfil the minimal standard, while lard does not meet the criteria of edible animal oils.

1. Introduction

The demand for meat has been growing rapidly in Indonesia. Consequently, it also generates high levels of waste and animal by-products such as bone, skin, and animal fats from processing plants. (Delgado *et al*., 2001; Sullivan and Diwyanto, 2007). In 2021, approximately 205 thousand tons of inedible animal fats are available annually. These animal fat by-products are generated from beef or mutton fat (tallow), pork (lard), chicken fat, and sheep or goat fats and are commonly used as raw material for oil production (Zhou *et al*., 2021).

Fat includes various fatty acids that have many biological functions (Strayer, 2006; Nielsen, 2006). Tallow and lard are the most important animal fats due to their broad range of applications in food, animal feeds, and in the manufacture of oleochemicals for several pharmaceutical and cosmeceutical products (Alvarez and Rodríguez, 2000). Generally, animal fats are extracted by the rendering process, a general method for animal oil preparation, which applies heat to allow the fat out of the adipocyte tissue (Lin and Tan, 2017). The rendering process is divided into two types, dry rendering (DR) and wet rendering (WR), depending on the water requirement (Sharma *et al*., 2013). A previous study indicated that the collection of animal fats and tallows through DR and WR methods suitably met all the criteria for physicochemical properties and fatty acid profiles (Chizoo *et al*., 2017; Limmatvapirat *et al*., 2021). Moreover, both methods were also suitable for lard oil for the food processing industry (Lee *et al*., 2018) and biodiesel production (Dias *et al*. 2009; Azike and Raji, 2019).

Indonesian local beef cattle and pigs have been well studied on the characteristics of phenotypic traits, polymorphism traits, production management, feeding methods, meat, and nutritional quality (Agus and Tri, 2018). Since the quality and characteristics of fats also depend on several factors, such as genetic type (species) (poultry: duck, goose, chicken) (Zanetti *et al*., 2010; Dal Basco *et al*., 2012; Boschetti *et al*., 2016), feed composition or methods (Bostami *et al*., 2017; Zaki *et al*., 2018), production system (Küçükyilmaz *et al*., 2012), and animal body parts (e.g., bone, skin, meat)

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(Straková *et al*., 2010; Taulescu *et al*., 2010), it is useful to gain information related to the quality attribute of local tallow and lard in Indonesia rendered by traditional methods. Concurrently, the world's scientific literature has also recently paid much attention to the composition of fatty acids in animal raw materials of various origins. As mentioned above, this study aimed to provide information on the physicochemical properties and content of fatty acid profiles of beef tallow and lard rendered to evaluate the impact of rendered fats using two different traditional methods. The extracted oils were evaluated for yields, color, acid value, saponification value, peroxide value, and fatty acid profiles.

2. Materials and methods

2.1 Materials and chemicals

Local beef and pig fats were collected from local slaughterhouses in Yogyakarta, Indonesia. The fats were cut off into small pieces, mixed thoroughly, and stowed in bags at -20°C until rendering was conducted. All chemicals used for determining fat quality were of analytical grade (Merck).

2.2 Extraction of beef tallow and lard

Previous studies showed that extracted fats were prepared by DR and WR methods using temperatures between 90 and 100°C (Ponphaiboon *et al*., 2018). For the WR method, beef fats and lard were separately weighed at 100 g along with water (1:2, w/w), while DR was performed without water in the rendering processes. Then, all samples placed in a beaker were rendered at temperatures between 90 - 100°C for 1 hr, which were thawed, cleaned, washed, and dried by absorbing the residual surface moisture on fat tissues. The clear oil was further filtrated using fine muslin and stored at 5°C after cooling to room temperature.

2.3 Physicochemical properties

The rendering yield (yield %) was calculated through the extracted oil weight (g)/fat sample weight (100 g) × 100. The samples were also analysed to determine their color intensity, acid value, saponification value and peroxide value (rancidity).

2.4 Color intensity

The yellow color intensities of oils were evaluated using a UV–Vis spectrophotometer, in which oils were placed and recorded in a 1 cm cuvette at 425 nm.

2.5 Acid value

The AOCS Official Method Cd 3d-63 was used to determine the acid value (AV) (American Oil Chemists' Society [AOCS], 2009). A total of 5 g of each oil sample was dissolved in 50 mL of ethanol solution (95%). The solution was titrated with a potassium hydroxide of 0.1 N in the presence of phenolphthalein. The AV is determined as mg of potassium hydroxide that neutralizes the free acids present in 1 g of oil. Data were calculated by employing AV (mg KOH/g oil) = mg of KOH to neutralize the FFA in 1 g of fat. The result was calculated using the formula AV (mg KOH/g oil) = (sample with oil – blank without oil) × 56.1 × N NaOH/ weight of sample used.

2.6 Saponification value

The SV was tested according to the AOCS Official Method Cd 3-25 (AOCS, 2009). Approximately 2 g of each oil sample was dissolved in 25 mL of alcoholic potassium hydroxide. The mixture was heated to saponify the oil by refluxing, and the unreacted potassium hydroxide was titrated with 0.5 N standard hydrochloric acid (HCl) solution using phenolphthalein as an indicator. A black sample was also prepared and titrated accordingly. The weight (mg) of potassium hydroxide needed to saponify 1 g of oil is expressed as the saponification value. The result was calculated using the formula SV (mg KOH/g oil) = (blank without oil – sample with oil) × 56.1 × N HCl/weight of sample used.

2.7 Peroxide value

The AOCS Official Method Cd 3d-63 was used to determine the peroxide value (PV) (AOCS, 2009). A total of 5 g of each oil sample was dissolved in 30 mL of a mixture of glacial acetic acids and chloroform at a ratio of 3:2, and then 0.5 mL of saturated potassium iodide solution was added. Approximately 30 mL of distilled water was added and thoroughly mixed after shaking for 1 min. This mixed solution was titrated with 0.01 N sodium thiosulfate to gain pale yellow, and a 2 mL starch indicator was added to reach the equivalent of colorless. The peroxide value is the milliequivalents of peroxide oxygen per 1 kg of oil. Data were calculated using the PV (meq O_2/kg oil) of S × M × 1000/g, where S = mL $Na_2S_2O_3$ (blank corrected) and M = molarity $Na_2S_2O_3$ solution.

2.8 Fatty acid profile

An automated liquid sampler and a Model 7890B Gas Chromatography Flame Ionization Detector (GC-FID) were utilized to analyse fatty acid composition. A capillary fused silica column DB-WAX (30 m × 250 µm × 0.25 µm) was operated under programmed conditions. A slope, starting, and final temperatures of 10°C, 190°C, and 280°C at a flow rate of 1.5 mL/min were used to study the column programming. The direct comparison

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of peak areas and retention times of fatty acid methyl esters (FAMEs) in oil samples with those collected from the CRM47885 FAME mix 37 Supelco standard helped in the identification and quantification of fatty acids.

2.9 Statistical analysis

All measurements were conducted in triplicate because they are represented as the mean ± standard deviation (SD). The results were analysed by employing multiple t-tests and the Holm–Sidak method (for statistical significance) in GraphPad 8.4.3 to determine the differences among yields and physicochemical properties. Additionally, a test was carried out to compare the fatty acid content between both methods in each species. Statistically, a P value of <0.05 was considered a significant difference. Two-dimensional principal component analysis (PCA) was assigned to discriminate the fatty acid profile of the oil from tallow and lard rendered using DR and WR methods. R-version 4.2.1 was used for PCA with the ggplot2 and ggfortify packages (R Core Team, 2018).

3. Results and discussion

3.1 Yield and sedimentation

The yield and sedimentation results are shown in Table 1. The mean yield value of the DR and WR methods was not significantly different between tallow and lard. Sharma *et al*. (2013) reported that both DR and WR methods are better for the recovery of fats. Zhang *et al*. (2013) explained that the temperature and duration of rendering affect the efficiency and quality of the final oil products. The difference in yield was probably because of different raw materials. However, sedimentation was found in all tallows rendered by DR and WR methods, except lard rendered by wet rendering methods.

3.2 Color intensity

The four rendered oils exhibited the yellow color of natural oil. The yellow color intensities of the oil from the DR tallow and lard (0.61±0.02 and 0.23±0.02, respectively) were higher than those from the WR tallow (0.39±0,03) and WR lard 0.07±0.01) methods, as shown in Table 1. A lower value indicated a bright yellow color and vice versa. Table 1 shows that WR for both tallow and lard had a brighter yellow color than DR oil. Sheu and Chen (2002) revealed that with increasing temperature levels or rendering duration, the oil product became darker and browner. In addition, Chumngoen and Tan (2015) also explained that the yellowness of oils determined their quality. In this study, two rendering methods provided significantly different results (P<0.05) of yellow color in the same row among each species.

3.3 Acid value

Table 1 illustrates that the WR method for both tallow and lard showed significantly higher acid values (1.72±0.07 and 1.68±0.11 mg KOH/g, respectively) than the DR method of tallow (0.67±0.11 mg KOH/g) and lard (0.79±0.11 mg KOH/g). A comparison between oil samples obtained by DR and WR methods provides significantly different results. However, all values did not exceed the limits of 2.5 mg KOH/g as suggested by the Codex Standard for Named Animal Fats CODEX-STAN 211-1999, FAO/WHO. The AV plays a role in the stability and oil flavour quality. A high temperature of the rendering method influences the production of free fatty acids in the oil, and it can stimulate the oxidation of oils (Ponphaiboon *et al*., 2018). Lin and Tan (2017) also indicated that water activity might influence the oxidation of fats.

3.4 Saponification value

This study shows no difference (P>0.05) in the SV tested in all samples. The saponification values were 201.03±0.81 mg KOH/g (DR) and 200.56±1.40 mg KOH/g (WR) in tallow rendered, while the lard rendered was obtained at 201.03±0.81 mg KOH/g (DR) and 200.56±1.40 mg KOH/g (WR), as indicated in Table 1. All the rendered samples were within the permissible limits set by the Codex Standard and FAO/WHO for Named Animal Fats CODEX STAN 211-1999 for edible fats and oils. Sharma *et al*. (2013) revealed that the saponification test helps in determining the average

Table 1. Physicochemical properties.

Parameters	Tallow		Lard	
	DR	WR	DR	WR
Yield (%)	80.68±0.58^{a}	81.33±0.58^{a}	70.67±0.58^{a}	70.67±0.58^{a}
Sedimentation (±)	+	+	+	-
Color (OD at 425 nm)	0.61±0.02^{a}	0.39±0.03^{b}	0.23±0.02^{a}	0.07±0.01^{b}
Acid value (mg KOH/g oil)	0.67±0.11^{b}	1.72±0.07^{a}	0.79±0.11^{b}	1.68±0.11^{a}
Saponification value (mg KOH/g)	200.56±1.40^{a}	201.03±0.81^{a}	201.03±0.81^{a}	200.56±1.40^{a}
Peroxide value (mEq O_2/kg)	6.00±0.20^{a}	5.87±0.31^{a}	52.53±2.40^{a}	52.40±2.12^{a}

Values are presents as mean±SD (n = 3). Values with different superscripts within the same row are statistically significantly different (p<0.05). DR: dry rendering, WR: wet rendering.

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molecular weight of fatty acids. The high molecular weight of fatty acids (lower SV) has a smaller number of carboxylic groups per unit mass than the higher SV. According to Mikulcová *et al*. (2017), all samples have the potential for cosmetic application.

3.5 Peroxide value

The peroxide values of DR and WR tallow (6±0.2 and 5.87±0.31 mEq O_2/kg, respectively) were lower than the permissible limit (≤10 mg mEq O_2 per kg oil), whilst DR and WR lard was 8.5 times higher than tallow (Table 1). The PV of tallow was below the permissible limit, while lard does not meet edible fat and oil standards. Sheu and Chen (2002) reported that different rendering methods did not significantly affect the peroxide value. In addition, fat species also contribute to the PV of oil products. Pereira *et al*. (1976) described that the level of temperature and oxygen during the rendering process was attributed to higher PV in the DR method when compared with the WR method. Furthermore, there is a positive correlation between the PV and the color intensity of all samples, in which low peroxide but high yellow color intensity might represent complete autooxidation in tallow, while higher peroxide value of lard showed partial lipid oxidation (light-yellow) occurred in the oil (Ponphaiboon *et al*., 2018).

3.6 Fatty acid profile

Table 2 shows the fatty acid compositions collected from tallow and lard using two distinct rendering methods. The DR method had a higher difference ($P<0.05$) in total saturated fatty acids (SFAs) than the WR method in all samples, whereas the WR method had higher ($P<0.05$) levels of total unsaturated (UFAs) and monounsaturated fatty acids (MUFAs) than the DR method. This study showed a strong relationship between the PV and the average SFAs content of all samples. It indicated that the increase of UFAs contributes to the PV that is caused by double bonds in the structure.

Although a few fatty acids were not observed in the lard rendered by the DR method, like pentadecanoic, margaric, and linolenic acids, this study shows that tallow and lard at least consist of major fatty acids, such as oleic acid, palmitic acid, and stearic acid. However, the fatty acid composition of all samples does not meet the acceptance criteria from the Codex Standard and FAO/WHO for Named Animal Fats CODEX STAN 211 -1999 for edible fats with oils. Previous studies emphasized that tallow and lard tend to be affected by factors such as sex, age, health status, genetic profile, diet, and freshness (Straková *et al*., 2010; Taulescu *et al*., 2010; Zanetti *et al*., 2010; Dal Basco *et al*., 2012; Küçükyilmaz *et al*., 2012; Boschetti *et al*., 2016; Bostami *et al*., 2017; Zaki *et al*., 2018).

3.7 Principal component analysis

The discrimination among treatment groups according to their fatty acid profile is clearly shown in Figure 1. The first two principal components (PCs) explained 98.96% of the variation. PC1 explains 89.89% of the variability, indicating that the data obtained are normally distributed and that the model is acceptable and corresponds with previous studies (Utama *et al*., 2018; Utama *et al*., 2022). The cluster pattern shows that

Table 2. Fatty acid (%) profile of Indonesia local tallow and lard obtained by two different render methods.

Fatty acids (g per 100 g sample)	Criteria of tallow*	Indonesia local tallow		Criteria of lard*	Indonesia local lard	
		DR	WR		DR	WR
C12:0 (lauric acid)	<0.5	nd	nd	<0.5	1.64±0.05^b	3.81±0.03^a
C14:0 (myristic acid)	2-6	5.09±0.12^a	4.81±0.06^b	1-2.5	3.68±0.03^b	5.96±0.05^a
C15:0 (pentadecanoic acid)	0.2-1	1.52±0.05^a	1.58±0.03^a	<0.2	nd	0.32±0.01
C16:0 (palmitic acid)	20-30	30.49±0.48^a	28.59±0.22^b	20-30	22.03±0.13^b	24.70±0.20^a
C17:0 (margaric acid)	0.5-2	2.16±0.05^b	2.34±0.04^a	<1	nd	0.27±0.02
C18:0 (stearic acid)	15-30	30.19±0.24^a	27.35±0.08^b	8-22	19.63±0.08^a	6.25±0.03^b
C18:1 *cis*-9 (oleic acid)	30-45	23.01±0.03^b	27.74±0.03^a	35-55	27.42±0.18^b	33.05±0.03^a
C18:2 *n*-6 (linoleic acid)	1-6	nd	nd	4-12	20.63±0.04^b	21.14±0.05^a
C18:3 *n*-3 (linolenic acid)	<1.5	nd	nd	<1.5	nd	1.81±0.06
Total saturated fatty acids (SFA)		69.46±0.86^a	64.55±0.22^b		46.98±0.16^a	41.28±0.26^b
Total unsaturated fatty acids (UFA)		30.54±0.86^b	35.45±0.22^a		53.02 ±0.16^b	58.72±0.26^a
Total monounsaturated fatty acids (MUFA)		23.01±0.03^b	27.71±0.03^a		27.42±0.18^b	33.05±0.03^a
Total polyunsaturated fatty acids (PUFA)		nd	nd		20.63±0.05^b	22.96±0.12^a

Values are presents as mean±SD (n = 3). Values with different superscripts within the same row are statistically significantly different ($p<0.05$). DR: dry rendering, WR: wet rendering, nd: not detected.

*Standard for Named Animal Fats (CXS 211-1999) by Codex Alimentarius (2023). Underlined values conform to the acceptable limit set.

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different rendering methods had a slight effect on the fatty acid profile of the tallow. On the other hand, rendering methods had a larger impact on the fatty acid profile of lard. The WR method reduced the proportion of stearic acid in tallow and lard, but the effect was much higher in lard. In this study, a higher proportion of stearic acid was found in beef tallow than in lard. This finding is in line with a previous study (Park *et al*., 2019). Therefore, the reduced amount of stearic acid in lard was much more significant than that in beef tallow. Both oils lost a significant amount of oleic acid in the DR method. The sensitivity of oleic acid against heat was also found in olive oil rendered from its mesocarp and intramuscular fat of beef (García-Inza *et al*., 2016; Utama *et al*., 2018). Stearic acid and linoleic acid were the distinguishing factors between tallow and lard according to the loading plots. Stearic acid was the predominant fatty acid found in tallow, while linoleic acid was found only in lard. Park *et al*. (2019) reported that the proportion of linoleic acid in lard was much higher than that in beef tallow. The PCA plot emphasizes the differences between two fat sources and two rendering methods.

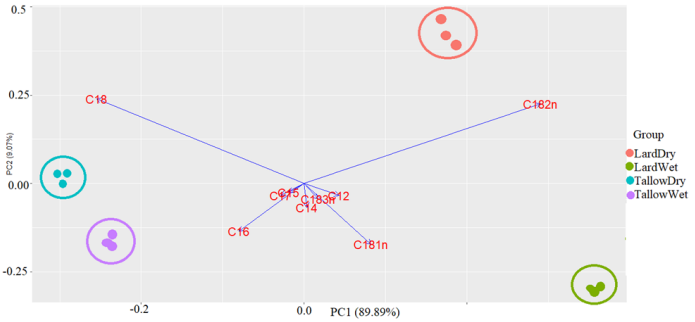

Figure 1. PCA plot showing the fatty acid loadings and differences in oil rendered from tallow and lard using DR and WR methods.

4. Conclusion

The physicochemical properties and fatty acid profiles of Indonesian local beef tallow and lard were reported and compared by two rendering methods. Both rendering methods showed the highest yield, while the yellow color intensity of oils from the DR method was higher than that of the WR method. The physicochemical properties of AV, SV, and PV of tallow collected from the DR and WR methods met the criteria for an international standard with the exception of lard. The number of each fatty acid of the two methods in each species was significantly different, although the major fatty acids existed (oleic acid, palmitic acid, and stearic acid). Tallow meets the limit for edible fat and oils according to physicochemical properties, while the fatty acid profile does not fulfil the limit requirement. Lard does not conform to the limits of animal oils in terms of its physicochemical properties and fatty acid profile.

Conflict of interest

No potential conflict of interest relevant to this article was reported.

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