Research Article

The Discriminant Method between Lard, Virgin Coconut Oil (VCO), Chicken and Beef Fat Based on Triglycerides Composition Using HPLC-UV

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ABSTRACT

A discriminant method as a model that can be used for identification of the lard in the foods. This research about the discriminant process between lard, VCO, chicken and beef using HPLC-UV. The results showed that the triglycerides composition in the lard and VCO have different. This result showed that the lard, VCO, chicken, and beef can be distinguished using principal component analysis. However, the analysis of triglycerides composition using HPLC-UV is not specific and simple method because of the other hydrolysis products from triglycerides will be detected with UV detector. The TGA analyses are very difficult to controlling the hydrolysis process. Then, the authentication and discrimination based triglycerides composition can't applicable using HPLC-UV instrument. This is new information for authentication halal food which the HPLC-UV instrument have a weakness for authentication analysis based on TGA composition. The solution for this problems is optimized the TGA hydrolysis process and the derivatization of fatty acid with 2.4 dibromo acetophenone.

Keywords: triglycerides composition, discriminant method, hydrolysis process, HPLC

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INTRODUCTION

Islam regulating for food labeling and the food was consumed by Moslems to giving the halal products. This regulation because the food ingredients were consumed can be classified in halal and haram [1]. Besides, the Islamic religion also considers to food composition as fat. The high fat content in the food will hold up the other substances such as vitamins, a nutrient which was needed for the body. However, people also need some kind of essential fatty acids such as linoleic acid (omega 3), omega 6, omega 9 which is very good for health. According to [2], that the ease of fatty acids can be digested by the body, it was depending on the chain length of fatty acid and position of fatty acids in the triglyceride molecules.

Recently, the development of methods for identification and characterization of fatty acid content from animal and vegetable fats. Reference [1] reported that increasingly the complex products will need to develop the analytical methods for authentication process the halal products. The development of identification methods for fat using FTIR combined PLS [3], PCR instrument [4-5] HPLC [6].

The discriminant process of pork, beef, chicken, mutton and chevon based on their primary amino acids content using HPLC-UV was be done [7]. This study for identification and discrimination the lard, VCO, Chicken and beef based on triglycerides composition. However, this research focused as an alternative method for a halal authentication process.

EXPERIMENTAL SECTION

Materials
Acetonitrile (Merck), Methanol (Merck), Hexane (Merck), Na_2SO_4 Saturated, VCO, Pork, Chicken, and Beef (Market in Yogyakarta)

Extraction Process of Triglyceride from Pork, Chicken and Beef
Two hundred gram samples were extracted with n-hexane using Soxhlet for 6 hours. The result of extract then added with Na2SO4, then filtered and evaporated to obtain the oil. The oil is stored at a temperature of 20 ^{o}C .

Analysis of Triglycerides Using HPLC UV
Ten mg fat from VCO and Lard are placed on the tube then   homogenized using a vortex. After that, the solution is diluted in a 50 ml flask with methanol. Finally, the solution was analyzed by HPLC (Shimadzu, LC 20 AT, Jepang) at a wavelength of 203 nm using a mobile phase composition of Acetonitrile : Methanol = 9 : 1.

Data Analysis
The analysis process for authentication halal was performed with processing the data of chromatogram of triglycerides from HPLC using chemometrics, especially PCA (Principal Component Analysis) using software The Unscrambler 10 (Camo software).

RESULTS AND DISCUSSION

The discriminant process between VCO, lard, chicken, and beef because of their triglycerides composition have different. According to [8] reported that the triglycerides compositions of coconut oil are 5 types of triglycerides, while Lard has 10 types of triglycerides. Reference [9] and [8] reported that the dominant of triglycerides composition from lard contains triglycerides such as palmitooleoolein (POO), palmitooleoolein (POS), and palmitooleopalmitin (POP). VCO has a dominant triglycerides were composed of medium chain saturated fatty acid [10]. The different of TGA composition from fat will give the different result on the chromatogram. The result showed that VCO, lard, chicken, and beef chromatograms are used for the discriminant process. The results of the processing of principal component analysis (PCA) showed that the lard, VCO, chicken, and beef can be distinguished (Figure 1).
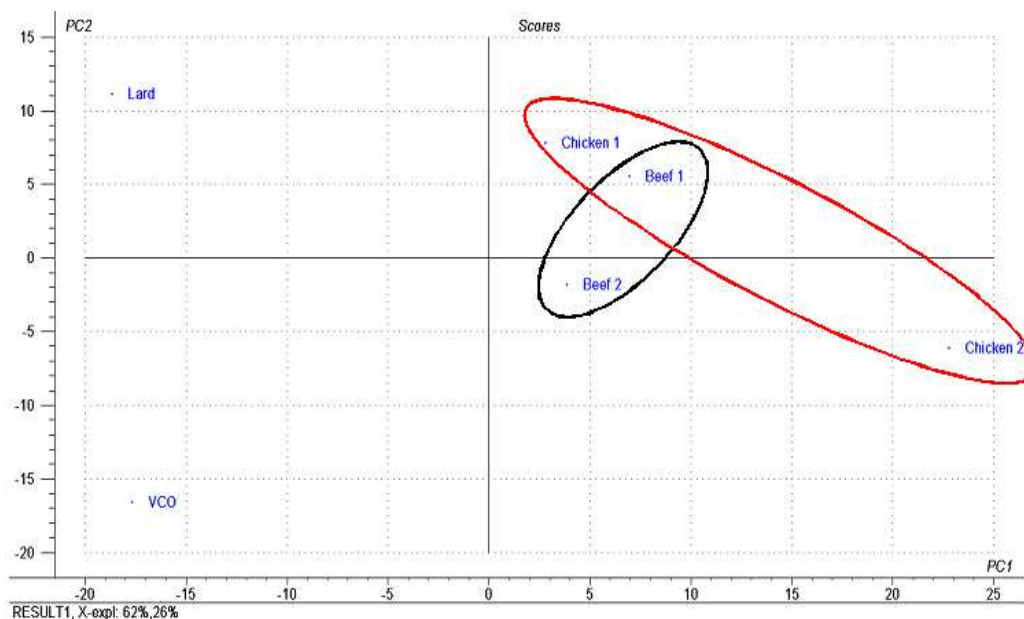

Figure 1. Results of data processing from HPLC-UV chromatograms of the Lard, VCO, Chicken, Beef Fat with Principal Component Analysis (PCA)

Figure 1. showed the discriminant process for all samples can be distinguished. The differences from triglycerides composition from lard, VCO, chicken and beef used as a basis in the discriminant process. The component of PC1 and PC2 indicated the value of 88 % in the discriminant process. This result indicates that there is approximately 12 % on the others PC. The distinction results with PC1 and PC2 components less from 95 %. However, the combined of PC1 and PC2 components have to be used in the differentiation process. The results also showed that the replication of measurements on chickens and cows are not precision. This is evidence that the chromatograms from HPLC-UV not only from the triglycerides but also from the hydrolysis triglycerides products. The triglycerides hydrolysis may occur in the fat storage process is done. Therefore, the analysis of the triglycerides composition using HPLC-UV has a problem because the TGA will be hydrolyzed to be other product. However, the discriminant process based on the triglycerides composition using HPLC-UV has problems in the controlling of hydrolysis process of triglycerides. Besides, the problems in the TGA analysis are the selectivity the UV detector because the TGA was analyzed on the

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lower of the maximum wavelength of 203 nm. The problems have indicated from the replicated analysis of TGA from chicken and beef is not precise. Reference [11] reported that the HPLC-UV can detect the TGA hydrolysis products such as methyl esters, fatty acids, monoglycerides, diglycerides. The hydrolysis process of TGA to be other product can refer to figure 2.
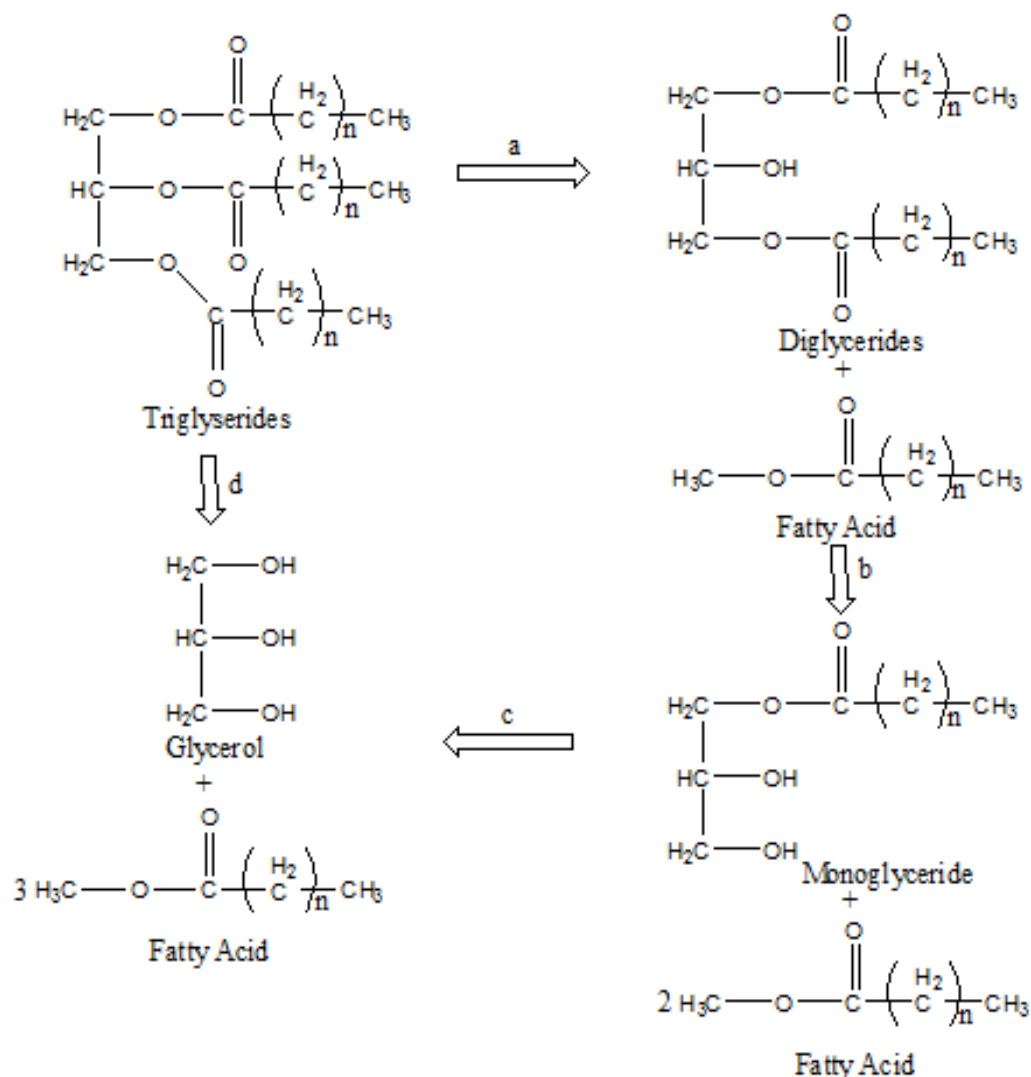
Figure 2. Triglyceride Hydrolysis Process: a) hydrolysis stage 1, b) hydrolysis stage 2, c) hydrolysis stage 3, d) Hydrolysis Total (The Complete Process of Hydrolysis)

The results of these triglyceride hydrolysis cause from replication analysis are not precise. The hydrolysis process of TGA cause the discrimination process is based triglyceride composition is not the simple and fast method. This is the reason for TGA composition on the samples must be hydrolyzed totally, and the discriminant process does not base on the TGA composition but base from the fatty acid composition. The results of discrimination processing of VCO, lard, chicken and beef also produce the large residual variance. This indicates showed that the discrimination result is generated each component has a large error (Figure 3). The solution for this problem is the discrimination process not based TGA composition but based on the fatty acids composition with optimized the derivatization of fatty acid with 2.4 dibromoacetophenone. This solution will give the discrimination process using HPLC-UV method is not the simple and fast method for authentication of the halal product.
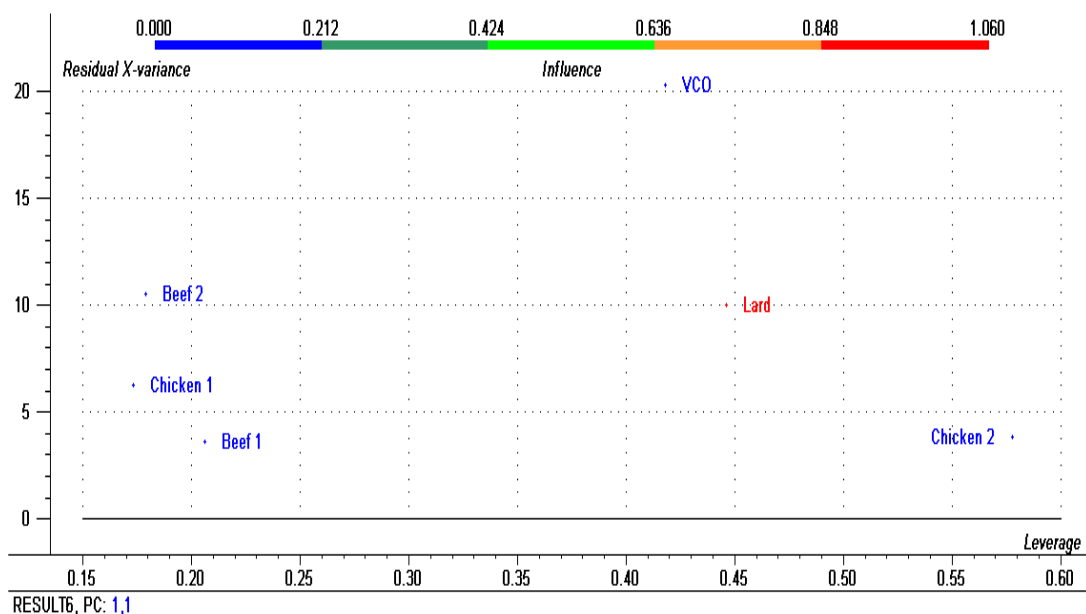
Figure 3. Results of residual x-variance for data processing from HPLC UV chromatograms of the Lard, VCO, Chicken, Beef Fat with Principal Component Analysis (PCA)

CONCLUSION

The results showed that the triglycerides composition in the VCO, lard, chicken, and beef can be distinguished. However, the analysis of triglyceride composition using HPLC-UV has problems such as maximum wavelength and the TGA hydrolysis. The UV detector is not specific for detecting TGA because it can detect the other hydrolysis product from TGA. The solution of these problems must to optimizing the hydrolysis process and its derivatization of fatty acid with 2.4 dibromoacetophenone.

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