Design of portable wireless impedance spectroscopy for sensing lard as adulterant in palm oil

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Abstract. This paper presents the design of portable wireless halal sensor using impedance spectroscopy technique for sensing lard as adulterant in palm oil. The impedance spectra of lard adulterated in palm oil (0.1\%, 0.5\%, 1\%, 5\% and 10\%) at the frequency range of 5 – 100 kHz at 45°C by using AD5933 Evaluation Board, a custom-built Interdigitated Electrode (IDE) and a Bluetooth module. The impedance spectra show that the binary mixture of lard and palm oil decreases as frequency increases. It is also found that the impedance spectra decrease as the concentration of lard adulteration in palm oil increase. PCA classification shows that different concentration of lard adulteration can be grouped in different cluster. PLS analysis shows high $R^2$ value, 0.97, indicating reliable prediction of the percentage of lard adulteration in palm oil.

1. Introduction

Adulteration is defined as the process of mixing two substances together. One of the most popular practices nowadays is the adulteration of lard into other food substances because lard is relatively one of the cheapest edible oils [1]. Lard is the term used to describe pig fat. Adulterating lard with other oils can reduce production cost.

Consumption of pork, lard or any pig’s derivatives is prohibited in Islam and Judaism. Because of this, system to detect adulterated lard in oils is needed to help address this issue. Several approaches have been investigated to detect lard. These methods can be grouped into two main groups which are labelling-based method and spectroscopy-based method. One example of labelling-based method is Polymerase Chain Reaction (PCR) technique [2].

Spectroscopy method uses the interaction of molecules with electromagnetic radiation (EMR) to study the physical and chemical properties of materials. Some example of spectroscopy methods included Electronic Nose (E-Nose) [3], liquid chromatography [4], Differential Scanning Calorimetry (DSC) [5], gas chromatography [6, 7], Fourier Transform Infrared (FTIR) spectroscopy [8, 9], dielectric spectroscopy [10, 11] and other electrical based spectroscopy techniques [12]. A number of impedance spectroscopy methods have been used in the study of impedance properties of oils like [12, 13]. In spite of that, the research on sensing lard as adulterant in palm oil is yet to be explored.

Interdigitated electrode (IDE) is cheap and small transducer, thus, widely employed together with the impedance measurement device. The sample’s impedance can be measured through the electrodes...
when connected to AD5933 Evaluation Board, a high precision impedance converter system. Lard and palm oil are two different materials that have different composition of fatty acids. It is expected that different concentration of lard adulterated in palm oil would cause different interaction of molecules and EMR. Hence a unique spectral behaviour can be used to sensing lard adulteration in palm oil.

This paper introduces the development of an electrical based spectroscopy technique using IDE. This wireless and portable impedance spectroscopy would provide a rapid in-situ halal verification system in the food manufacturing sector.

2. Materials and Methods

This section discusses the materials and methods used in designing portable wireless impedance spectroscopy for sensing lard adulterant in palm oil. First the hardware and software of the system is implemented so the measurement can be done using the system.

2.1. Hardware and software implementation

The sensor system consists of an IDE, Maker Uno microcontroller board, AD5933 evaluation board, HC-06 Bluetooth module and a mobile application.

The IDE is connected to AD5933 evaluation board and is used as the contact surface to measure binary mixture oil’s impedance. The value measured is processed by Maker Uno microcontroller board and sent to mobile application via HC-06 Bluetooth module. The IDE design was according to [13] with 0.5mm width, 0.43mm spacing and 16 teeth. The IDE pattern is designed using AutoCAD software and is fabricated at Department of Electrical and Electronics Engineering PCB (Printed Circuit Board) Lab, UPM.

![Interdigitated electrode drawing in AutoCAD.](image)

SCL (clock line) and SDA (data line) pins of AD5933 Evaluation Board are connected to Maker Uno A5 and A4 analog pin respectively, to retrieve the impedance values measured. Since no existing port is available in this board, the two connecting jumpers are soldered directly to the pins where SCL and SDA data is processed.

AD5933 chip calculates the sample’s impedance using the following formula

\[
\text{Impedance} = \frac{1}{\text{gain factor} \times \text{magnitude}}
\]  

(1)

The gain factor is to be set by conducting gain factor calibration. Calibration is important to ensure accuracy and reduce error during the experiment. For this purpose, a 4.7 kΩ resistor is used as a known impedance value. The 4.7 kΩ resistor is connected to Z pin of the AD5933 board, in which the IDE is connecting to during sensing. To further reduce the probability of error, the jumper that connects the AD5933 Evaluation Board and the resistor is set to be almost the same length with the jumper used for IDE.

Based on Lizhi [14], it is stated that the gain varies as frequency varies. The gain is also different at different temperature. Therefore, the calibration is conducted at every 5 kHz value between 5 kHz to
100 kHz. The calibration is done at 45°C in the temperature chamber. Initial gain is assumed to be 1 and magnitude is displayed in the mobile application. Frequency sweep is conducted 4 times and average value of magnitude is recorded for each frequency.

HC-06 Bluetooth module is connected to Maker Uno through pin 10 and 11, namely the transmitter (Tx) and receiver (Rx) pin. Maker Uno sends data in JavaScript Object Notation (JSON) data format to the 4-pin HC-06 Bluetooth module. The data is displayed on the mobile phone as output.

The software design involved is a mobile application that is used to display results from the lard detection system. Mobile application is chosen because almost everybody owns a mobile phone nowadays. The mobile application is developed using Android Studio, and is targeted for users with Android 5.0 and above. An open-source Bluetooth library is used for building an application with Bluetooth connectivity.

2.2. Sample preparation
Palm oil was bought off shelf from local supermarket. Adipose tissues of pig were rendered to extract the fats according to the method proposed by Marikkar [15]. The adipose tissues were collected from local markets. Melted fats were then filtered through double-folded muslin cloth to remove impurities. The collected lard was filtered through Whatman No.4 filter paper and a small proportion of anhydrous sodium sulphate (Na2SO4) to remove moisture.

Extracted lard was mixed in proportions ranging from 0.1 to 1% lard, in 0.5% increments, 1 to 10% lard, in 5% increments (w/w). A total of 5 blends were prepared - 99.9:0.1, 99.5:0.5, 99:1, 95:5, 90:10 (w/w) – and these were identified by the mass ratio of palm oil to lard.

2.3. Oil impedance measurement
The impedance of oil samples were measured using the custom-built IDE sensor connected to AD5933 Evaluation Board. The AD5933 Evaluation Board was calibrated at each frequency. Measurements were carried out at frequencies 5 kHz to 100 kHz with interval of 5 kHz. The samples were measured at 45 ± 0.1°C in a temperature controlled chamber (Espec SU221, Michigan, USA) as shown in Figure 3 to make sure all the samples were homogenous.

To ensure that only type of samples and lard concentration are the manipulative variable in this experiment, all other factors such as size of beaker, position of beaker, position of IDE inside the beaker, area of IDE that is submerged into the beaker and other interference factors are taken into account.

![Figure 2. Experiment flowchart.](image-url)

To measure impedance, the IDE is submerged into oil sample and sweep is started using mobile application. The frequency and impedance values are recorded in .csv file in the mobile application.
2.4. Statistical analysis
All the measurements were carried out in triplicate and mean values and standard deviations were calculated. Repeatability was assessed using relative standard deviation (%RSD) of triplicate measurement. Analysis of variance (ANOVA) at 5% level of significance was performed to compare the mean of samples between different frequencies. The Unscrambler software version 10.3 (CAMO Software AS, Trondheim, Norway) was used to carry out Principal Component Analysis (PCA) classification and Partial Least Square (PLS) analysis of the experimental data.

3. Results and Discussion
This section discusses the outcome from the design of the system and is divided into 4 parts, the outcome of the device design, the analysis of the data from impedance measurement, the PCA classification model and the PLS calibration model.

3.1. Wireless impedance spectroscopy platform
The mobile application that can be connected to Bluetooth and received data from Arduino is developed. The application consists of 2 pages, which are to select device page and home page as shown in Figure 4.

Figure 4. (a) Select device page, (b) Home page when no device is connected, (c) Home page when device is connected and data is received from AD5933 Evaluation Board, and (d) Data is saved into .csv file.
The system designed was able to take input from the IDE designed, process them by the Maker Uno and send the data to mobile application via HC-06 Bluetooth module. The data collected was successfully saved into .csv file and can be downloaded for data analysis as shown in Figure 5.

![Figure 5. Impedance data in the .csv file.](image)

### 3.2. Impedance properties of binary mixture of oil

Repeatability was evaluated at 3 measurements for each sample. The RSD values for 3 measurements were < 2%. This data indicate that the method is highly reproducible. In the frequency range of 5 – 100 kHz, the impedance spectra of binary mixture of oils indicated a significant decrease (p < 0.05) as shown in Figure 6.

The impedance spectra of binary mixture of palm oil decreased with increasing concentration of adulterant (lard) over the measured frequency range. The increment was more apparent at higher range of frequency, 70 – 100 kHz, as shown in Figure 6. The impedance spectra of lard were lower than that of palm oil, thus increasing concentration of lard in palm oil causes the spectra to decrease. However, the concentration 0.1% and 0.5% was observed to be difficult to be distinguished. Since discriminating adulteration of lard in palm oil directly by impedance is difficult, further investigation using PLS analysis is needed.

### 3.3. PCA classification model

Discrimination of palm oil adulterated with lard was carried out using PCA and PLS models. Figure 7 demonstrate the PCA score plot of palm oil adulterated with lard describing the projection of samples defined by the first (PC1) and second (PC2) components. PC1 accounts for the most variation in impedance spectra, while PC2 accounts for the next largest variation. PC1 accounted for 89% of the variation, while PC2 contributed to 7% of the variation, making up of 96% of variance for PC1 and PC2.
Figure 6. Impedance values of palm oil at different concentrations compared to impedance of pure lard.

Figure 7. PCA score plot for different concentration of lard in palm oil.
The observation of PCA score plot for all oil samples showed clearer and better performance in distinguishing different concentrations of adulterant from 0% to 100% (w/w). Each of the oil samples could be easily grouped in different clusters using impedance spectra.

3.4. PLS calibration model

The relationship between concentration of lard adulterant in palm oil and frequency of the impedance spectra were further assessed using PLS analysis. In this analysis, independent variables, X, were impedance of oils measured over the measured frequencies (20 frequencies), while the dependent variable, Y, was the concentration of lard adulterant in palm oil (%). The prediction capability of the regression model was assessed by cross-validation, i.e., the coefficient of determination ($R^2$) and standard error of root mean square (RMSE).

![Figure 8. Correlation loading plot of binary mixed oil samples.](image)

The correlation loading plot in Figure 8, displays information about the variables in the PLS model. The plot shows that variables from impedance spectra have an extreme position to the left of the plot along PC1, while variable concentration of lard adulterant was on the extreme opposite position to the impedance spectra along PC1, indicating a strong negative correlation to each other. This explains the relation where the impedance spectra decrease when the concentration of lard adulterant in palm oil is increases.
Figure 9. Prediction versus reference for PLS calibration model.

Figure 9 shows the regression model provided reliable prediction of the percentage of lard adulteration in palm oil. The $R^2$ was 0.97 and RMSE 6.19. From this result, impedance spectroscopy could be applied in predicting the concentration of lard adulteration in palm oil.

4. Conclusion
A portable wireless impedance spectroscopy was introduced for sensing lard adulterant in palm oil. The system was developed by using IDE, Maker Uno microcontroller board, AD5933 evaluation board, a Bluetooth module and a mobile application. The impedance spectra of binary mixture of lard in palm oil decreased with increasing concentration of lard adulteration over the measured frequency range. PCA classification plots for all samples showed clear performance in the distinguishing different concentrations of lard adulterated oil. Each of the samples could be easily grouped in different clusters using impedance spectra. PLS calibration model showed a good prediction capability for different concentration of lard adulteration in palm oil. Further investigation on distinguishing palm oil adulterated with lard using multivariate analysis is needed. Impedance spectroscopy could be beneficial in the quantitative determination of levels of lard adulterants in palm oil.

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