Effect of Sterilization Process on Deterioration of Bleachability Index (DOBI) of Crude Palm Oil (CPO) Extracted from Different Degree of Oil Palm Ripeness

Junaidah Mat Jusoh, Norizzah Abd Rashid, and Zaliha Omar

***Abstract*—Oil palm Fresh Fruit Bunch (FFB) sterilization process was studied using different degree of FFB ripeness (i.e. underripe, ripe, overripe) and loose fruits. The experimental runs were conducted using laboratory scale sterilizer at varied process condition. With application of Response Surface Methodology (RSM), the obtained results were used for analyzing interrelation between heating parameters and Deterioration of Bleachability Index (DOBI) of Crude Palm Oil (CPO) and performing sterilization process optimization. Both temperature and time were found to have influence on the DOBI of extracted CPO. However, sterilization time was found exerted greater influence as compared to temperature. By using numerical optimization method, optimum sterilization conditions with corresponding to DOBI were determined.**

***Index Terms*—DOBI, FFB ripeness, optimum FFB sterilization condition.**

I. Introduction

The importance of Crude Palm Oil (CPO) quality cannot be overlook due to the fact that good-quality refined oils cannot be produced from poor quality CPO [1]. It is essential to any palm oil mill to extract CPO with excellent quality and stability, particularly on oxidation [2]. Lipid oxidation in CPO is a process by which the unsaturated fatty acids in CPO react with oxygen from the air, resulting in rancidity [3] that might lead to poor oil bleachability and keepability. As a result, flavors and shelf life of the final food products will be degraded. Such degradation can be easily detected by the palate even at very low concentrations [3]. Deterioration of Bleachability Index (DOBI) reading is one of compulsory quality parameter used for measuring the oxidation level [2]. It has been officially added to the Malaysian Domestic Sales of CPO Contract since July 2004 [2].

Oil palm Fresh Fruit Bunches (FFB) and its loose fruits are two main groups of oil palm products used in CPO extraction process [5]. Mills usually will purchase loose fruits at higher prices than FFB due to its potential in contributing to higher Oil Extraction Rate (OER) [4], [5]. As been established by Malaysian Palm Oil Board (MPOB), FFB ripeness is classified into five main classes, namely unripe, underripe, ripe, overripe and rotten. Practically, palm oil mills continuously receive different degree of FFB ripeness in their daily operation though it is a compulsory for any oil palm estate to send minimum of 95% FFB ripeness percentage for maximum recovery with standard quality of the produced CPO. Plus, majority of the mills do not employ either segregation practice or stringent ripeness control onto the received FFB [5]. Thus, quantitative information obtained from studies that able to produce CPO from individual degree of FFB ripeness is clearly necessary since it might provide additional values to the current palm oil industry practice.

In the beginning of CPO extraction process, FFB will be subjected to sterilization process to accomplish numerous targets, especially for deactivating the biological factors that responsible for quality deterioration, as well as loosen the fruits in FFB for maximum fruit recovery during stripping and threshing processes [7], [8]. Since it is the earliest stage during the process, it ensures the success of other subsequent stages. There are few issues arisen during the sterilization process such as high oxidation risks and over sterilization that are said as leading to poor bleachability to the extracted CPO. These problems support the finding made by [9] which reported that CPO quality is greatly influenced by heating parameters of the process. Precise study for improving CPO quality with special attention to the FFB sterilization process therefore should be conducted. Though enormous work has been done to date regarding the CPO quality attributes, the interrelation between sterilization process and FFB ripeness and DOBI has not been thoroughly studied.

Response Surface Methodology (RSM) enables evaluation of interactive effects between the process factors and responses variables [10]. It is less laborious and time-consuming as it reduces number of experimental runs needed to provide sufficient information for statistically acceptable results. The main objectives of this study are to investigate the relationship between DOBI and individual degree of FFB ripeness, and determine optimum operating condition of FFB sterilization process at different degree of ripeness which satisfies the DOBI of the extracted CPO.

II. Materials and Methods

A. Raw Materials

The FFB were obtained from a local palm oil mill situated in Malacca, Malaysia. The samplings were performed according to MPOB FFB Grading Manual [11] with the assistance from certified mill FFB grader. Different batches of fruits had to be taken as the overall experiments were performed for a long period. The selected FFB afterward were carefully chopped into smaller form called spikelet to cope with utilization of laboratory-scale sterilizer.

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B. Experimental Design

With an aid of Design-Expert Version 8.0.1 software (Stat-Ease Inc., Minneapolis, USA), two-variable Central Composite Rotatable Design (CCRD) was employed to study the effect of FFB sterilization condition at different degree of ripeness on the response, namely DOBI, $Y_{1\text{-}4}$ (Y_1, underripe; Y_2, ripe; Y_3, overripe; Y_4, loose fruits). The independent operating variables were heating parameters of the process; sterilization temperature and time, X_1 and X_2, which varying between 100 to 120°C and 20 to 80 min, respectively (refer Table I). Five replicates run at the centre point were performed to allow the estimation of pure error. Accordingly, 13 experimental runs were generated for each degree of FFB ripeness. The experiments were conducted in randomized order to minimize the effects of unexplained variability in the observed response due to extraneous factors.

TABLE I: EXPERIMENTAL RANGE AND LEVELS (CODED AND ACTUAL) OF THE INDEPENDENT VARIABLES (PROCESS FACTORS)

Variables	Range and Levels				
	Lowest	Low	Centre	High	Highest
	-1.414	-1	0	+1	+1.414
X_1, (°C)	95.86	100	110	120	124.14
X_2, (min)	7.57	20	50	80	92.43

C. FFB Sterilization

For each experimental run, 20 spikelets (~3 kg) were placed in swiftlock programmable autoclave, a laboratory-scale sterilizer (Astell Acientific, 5000 series, UK) and subjected to a sterilization process according to the generated experimental runs. The sterilized spikelets then were subjected to extraction process.

D. In Laboratory CPO Extraction

The spikelets were detached from the stalk and the mesocarp was manually peeled off from the nut using stainless steel blade. The peeled mesocarp then was submerged (5-10 minutes) into boiling water before it was pressed using a coconut milk presser (bought from the local market) to facilitate oil extraction. The obtained mixture was transferred into separating funnel with surrounding temperature at approximately 60°C to aid oil clarification process. After 45 min, three distinctive layers of oil, water and sludge were formed at the upper, middle and bottom part of the funnel, respectively. The clarified crude oils were then subjected to purification process right after water and sludge layers carefully removed from the funnel. The process was performed using centrifuge (Sorvall® RC 26 Plus, Kendo Laboratory, USA) at 10000 rpm and 45°C for 10 minutes. Finally, the purified CPO was subjected to drying process using vacuum oven (15inHg max; 80°C) for 150 min. The extracted CPO was instantly subjected to DOBI analysis, without any prior storage.

E. Deterioration of Bleachability Index (DOBI) Analysis

DOBI is a numeric ratio of carotene (pro-Vitamin A) and secondary oxidation. Dividing the two gives an indirect net result of oxidation that is amplified. Determination of DOBI was carried out according to MPOB Test Methods [12]. Three replicates of the analysis were performed for every extracted CPO, and the reported value is the average of the three values.

F. Response Surface Analysis

The measured data was analyzed using the Design-Expert Version 8.0.1 software. The most appropriate polynomial regression models were chosen for predicting the interactive effects of heating parameters (X_1 and X_2) of sterilization on the response ($Y_{1\text{-}4}$). The fitness of each developed model was evaluated by the Analysis of Variance (ANOVA). The fitted models were also interpreted into three-dimensional graphs (response surface plots) by assistance of Statistica 10.1 software to ease the model evaluation.

G. Multiple Response Optimization

Based on the results obtained from the models analyses, numerical optimization method has been used for determining the optimum condition of each sterilization operation at different degree of FFB ripeness. The desired goals for each variables and response were chosen. Accordingly, the X_1 and X_2 were kept within the study range while the $Y_{1\text{-}4}$ was set to be maximized, as good CPO quality was indicated by high DOBI value. In order to search a solution maximizing the response, the goal is combined into an overall composite function, $D(x)$, called as the desirability function [13].

III. RESULTS AND DISCUSSION

A. Experimental Data, Model Fitting and Statistical Analysis

The measured DOBI value throughout this study ranged from 0.99 to 6.69, with minimum and maximum values were recorded from CPO extracted from ripe and overripe FFB, respectively. The minimum-to-maximum ranges obtained from overall experimental runs were represented in Fig. 1.

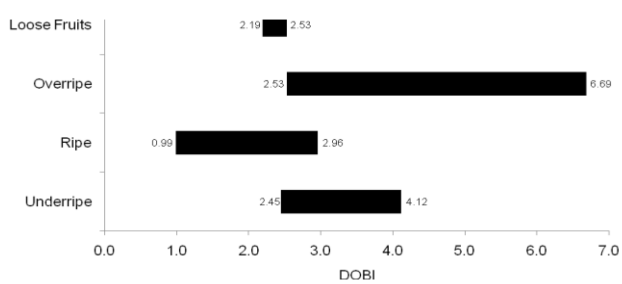

Fig. 1. DOBI value (minimum-to-maximum range) for CPO extracted from different degree of FFB ripeness.

The minimum-to-maximum ranges shown in Fig. 1 indicate that each individual degree of FFB ripeness was able to produce fairly good DOBI grade CPO [1]. The recorded data also shows that individual degree of FFB ripeness was conformed to the specification for production of Malaysian Standard Quality Grade II CPO (MS 814:2007), i.e. minimum 2.2 of DOBI value [14]. Surprisingly, no apparent data trend could be observed from Fig. 1. However, overripe FFB has recorded the highest DOBI value, followed by underripe, ripe and loose fruits. This might be due to the rich and deep orange-red color usually found in overripe oil palm fruits, indicating extremely high carotene content [15]. Loose fruits were found recorded the narrowest DOBI range (i.e. 2.19 to 2.53). This is probably owing to its similarities of physical- and chemical- characteristics between individual loose fruit since none of them were attached to the bunch, unlike any other samples.

Power law transformation usually should be considered before experimental data were fitted into any regression model [16]. Based on the results, the power law transformations were unnecessary since the max. to min. ratio were less than ten [10], i.e 1.686, 2.989, 2.644 and 1.153 for underripe, ripe, overripe and loose fruit, respectively. The experimental results therefore were directly subjected to model fitting where the polynomial regression models will sufficiently explain the effect of sterilization process on DOBI of CPO. Table II shows the Fit Summary report of models, which summarizing results of Sequential Model Sum of Squares (SS) and Lack-of-Fit (LOF) tests, including regression coefficient (R^2) values as reported in Model Summary Statistics section in the software.

A statistical model is developed through combination of terms for estimating factor effects. Three different polynomial models were listed and analyzed in Table II for model fitting purposes. The cubic models were negligible due to insufficient of experimental runs for independently estimating all the terms in the model.

TABLE II: FIT SUMMARY REPORT FOR FITTING MODELS

Resp.	Source	SS	LOF	Model Summary Statistics		
		p-value	*p*-value	R^2	Adj. R^2	Pred. R^2
Y_1	LNR	<0.0001	0.3791	0.8850	0.8621	0.7846
	2FI	0.3032	0.3765	0.8985	0.8647	0.7069
	QRC	0.8395	0.2312	0.9034	0.8345	0.5158
Y_2	LNR	<0.0001	0.0021	0.9183	0.9020	0.8217
	2FI	<0.0001	0.0669	0.9884	0.9845	0.9642
	QRC	0.4679	0.0479	0.9907	0.9840	0.9421
Y_3	LNR	0.0002	0.0415	0.8145	0.7774	0.6135
	2FI	0.8810	0.0314	0.8150	0.7533	0.5982
	QRC	0.0008	0.6462	0.9762	0.9592	0.9216
Y_4	LNR	0.0536	0.0087	0.4431	0.3317	-0.2004
	2FI	0.0003	0.1136	0.8768	0.8358	0.6358
	QRC	0.5771	0.0746	0.8947	0.8195	0.3724

Note: LNR= Linear; 2FI= Two Factor Interaction; QRC= Quadratic

The sequential model SS was analyzed for testing the hypotheses of model parameters. The ultimate aim of sequential analysis is to select the highest degree of non-aliased model that has a p-value of additional terms that is lower than the chosen level of significance, i.e. 0.05 [10]. The p-value (Prob.>F) approach was used for the testing. If the p-value is very small, it rejects the null hypothesis, H_0 (there is no factor effect) and thus concludes that at least one of the two process parameters, i.e. X_1 (temperature) and X_2 (time) has a non-zero regression coefficient in the developed model.

The LOF tests' results are useful for diagnosing how well each of the full models fits the data [16]. If the p-value obtained from this test is very small (less than 0.05), then LOF is significant. This will only discredit the fitted models as the variation in the model points significantly differs from the variation in the replicated points. The selected model is supposed to have insignificant LOF, i.e. *p*- value for LOF to be greater than 0.10.

Regression Coefficient, R^2 is a measure of the amount of variation around the mean explained by the model, which also known as a degree of fit measurement that is beneficial for measuring the proportion of total variability explained by the model. The best R^2 value for a good model fitting is somewhat that is closer to 1.0 with not less than 0.8 [10]. A value of 1.0 represents the ideal case at which 100 percent of the variation in the observed values can be explained by the chosen model. Predicted- and Adjusted- R^2 should be within 0.20 of each other. Otherwise, there may be a problem with either the data or the model [16].

For underripe FFB, extremely low p-value of <0.0001 indicates that adding linear terms (X_1 and X_2) to the intercept has significantly improved the model fit. Linear model showed insignificance of LOF with highest p-value, i.e. 0.3791, as compared to 2FI and quadratic models with 0.3765 and 0.2312, respectively. The R^2 values of available models for underripe FFB are 0.8850, 0.8985 and 0.9034 for linear, 2FI and quadratic models, respectively. Linear model was considered for representing the fitted response values since the difference between the Adjusted- and Predicted- R^2 of quadratic model was exceeding 0.2, i.e. 0.3187, as compared to linear and 2FI, with 0.0775 and 0.1578, respectively.

Both linear and 2FI models of ripe FFB showed extremely low of *p*-value, i.e. <0.0001 in sequential analysis. All models were found to have relatively poor p-value in LOF test with 0.0021, 0.0669 and 0.0479 for linear, 2FI and quadratic, respectively. 2FI were chosen for model fitting since 2FI model is higher degree of polynomial model as compared to linear model. Though the p-value of 2FI model in LOF was considered quite low, it exceeded 0.05 than the linear model p-value that was extremely low and unacceptable. All three models in ripe FFB have high R^2 values with 0.9183, 0.9884 and 0.9907 for linear, 2FI and quadratic models, respectively. For a case like this, it is important to bear in mind that R^2 will always increase as the variable is added to the model, regardless of whether the additional variable is statistically significant or not [10]. It is still possible for models that have large values of R^2 to yield poor predictions of new observations or estimates of the mean response. Therefore, the 2FI model was suggested as it was the highest degree of the polynomial models that showed high value of R^2 with significance in model sequential analysis and insignificance of LOF test.

During sequential analysis of overripe FFB, both linear and quadratic models were found significant with p-value of 0.0002 and 0.0008, respectively. However, quadratic was the only model that has insignificance of LOF with p-value of 0.6462. As it is also a higher degree of polynomial model, quadratic should be chosen for model fitting. Furthermore, the quadratic model also recorded the highest value of R^2, 0.9762 as compared to the other models, i.e. 0.8145 and 0.8150 for linear and 2FI models, respectively.

Based on the loose fruits' result, it was easy to choose 2FI model for predicting the response behavior within the experimental region. In model sequential analysis, 2FI model was the only significant model with extremely low p-value, i.e. 0.0003. LOF test also revealed that 2FI model was also the only model that has insignificance of LOF with p-value of 0.1136. Though both 2FI and quadratic models were considered of having great values of R^2, Adjusted- and Predicted- R^2, 2FI was selected due to its consistent performance in model sequential analysis and LOF test.

Once the selected models were fitted, ANOVA were performed onto individual fitted models to examine the statistical significance of the model terms. The estimated coefficients values of the fitted models and statistical data obtained from ANOVA have been summarized in Table III. All of the fitted models, i.e. linear for underripe FFB (Y_1), 2FI for ripe FFB and loose fruits (Y_2 and Y_4), and quadratic for overripe FFB (Y_3) were highly significance models as they have extremely low of p-values with <0.0001 (Y_1, Y_2 and Y_3) and 0.0002 (for Y_4). The models' residual diagnostic plots were also generated to check goodness of models' fit. As expected, each of the fitted models satisfied the assumptions of ANOVA.

TABLE III: ESTIMATED COEFFICIENT VALUES (CODED TERMS) AND STATISTICAL DATA OBTAINED FROM ANOVA

Coefficients	Estimated Coefficient Values			
	Y_1	Y_2	Y_3	Y_4
Intercept	3.205	1.864	4.050	2.306
X_1	-0.113	-0.265	0.122	-0.053
X_2	0.489	-0.645	1.367	0.040
X_1^2	-	-	-0.539	-
X_2^2	-	-	0.299	-
$X_1 X_2$	-	0.273	0.048	-0.093
Model (p-value)	<0.0001	<0.0001	<0.0001	0.0002
Standard Deviation	0.160	0.074	0.250	0.033
Adequate Precision	17.795	51.079	24.705	15.952

The models also found with relatively low Standard Deviation, high Adequate Precision as well as achieving reasonable agreement between Adjusted- and Predicted- R^2. Adequate Precision measures the signal to noise ratio. If the ratio is found as greater than four, it indicates an adequate signal and the particular model can be used to navigate the design space [16].

A. Effect of Sterilization Process on DOBI of CPO Extracted from Different Degree of Oil Palm FFB Ripeness

Fig. 2 a) to d) illustrate the relationship between independent and dependent variables in three-dimensional representation of response surfaces generated by the fitted models equations. These figures represent DOBI of CPO as a function of sterilization temperature and time, which are useful for understanding the main effects of time and temperature of sterilization on the DOBI of CPO extracted from different degree of FFB ripeness.

As can be observed in Fig. 2 (a) to (d), linear coefficient for processing time (X_1) was found dominant in all of the fitted models. All of X_1 model terms were found to be highly significant with extremely low *p*-value of <0.0001 (for underripe, ripe and overripe FFBs) and 0.0077 (for loose fruits). Thus, it indicates that there was a strong and significant effect of the linear factor of time on the DOBI. This result approving previous finding made by [9] that the change in processing temperature seemed to be less important than the change in processing time.

Though time was considered as a major factor determining DOBI value, it should be noted that as the oil palm fruits were ripen, both X_1 and X_2 started to play equal role in affecting DOBI. ANOVA result also showed that for ripe FFB, both linear model terms (X_1, and X_2) and interaction term (X_1X_2) was found highly significant with p-value of <0.0001. As the time duration was increased, DOBI was found significantly decreased from 4.5 to 0.5, which might due to oil oxidation effect as a result of the lengthened thermal treatment [7]. The response surface also indicates the possibility of reaching greater DOBI value (approximately 5) if lower time and temperature were applied during the FFB sterilization process. This finding is in agreement with the works reported by previous researchers [9] that the good bleachability necessitates a proper air-release prior to sterilization, the shortest possible sterilizing time and lowest possible process temperature. However, it is impractical to use the combination of low time and temperature for sterilization process since the enzymatic fat splitting activity would only ceases at 55 ℃ [7]. Furthermore, shorter process time might also interrupt the stripping process as well as oil palm fiber detachment during digestion process later.

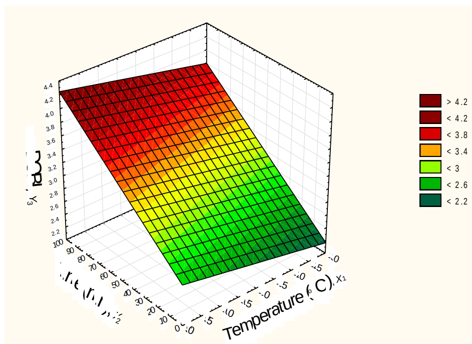

Fig. 2. a) Underripe FFB, Y_1.

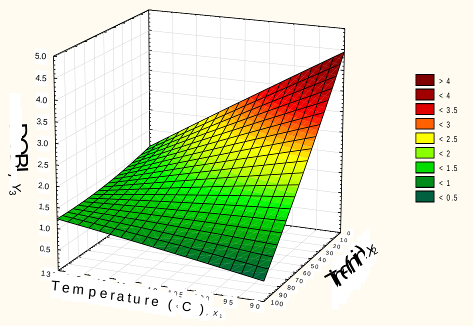

Fig. 2. b) Ripe FFB, Y_2.

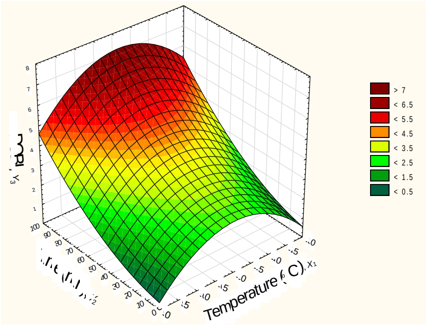

Fig. 2. c) Overripe FFB, Y_3.

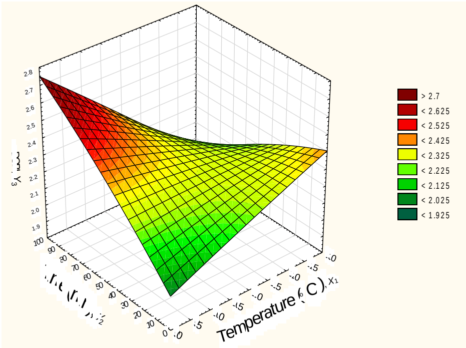

Fig. 2. d) Loose Fruits, Y_4.

Fig. 2. a) to d) Three-dimensional response for the effects sterilization process on DOBI of CPO extracted from different degree of oil palm FFB ripeness.

Quite expectedly, the DOBI of CPO extracted from loose fruits did not able to achieve minimum of satisfactory quality. As was outlined by Malaysian Palm Oil Council (MPOC), the good DOBI grade for CPO is 2.99 to 3.24. Unfortunately, if referred to Fig. 2d), maximum DOBI value that probably could be achieved by loose fruits' CPO was still low. The major reason of having such poor DOBI value was likely due to the very high secondary oxidation products that might exist in loose fruits, due to its physical condition that were naturally detached from its bunch. Thus, though it probably contains high carotene content, the DOBI value still could be very low. For a case like this, nothing much could be done by mill operation, except by avoiding excessive handling and reducing heavy metal contamination, especially copper and iron as they are classified as strong pro-oxidants [3].

B. *Sterilization Process Optimization*

Table IV shows the optimum condition of FFB sterilization process with the predicted values of DOBI. Though several solutions were suggested by the software, only solutions which possess maximum desirability value were selected (for each FFB ripeness) as optimum condition of FFB sterilization. The overall desirability score were recorded as positive value, which are desirable [16]. The optimum temperature suggested by the software was within lower temperature range. The suggested optimum time was 20 min for underripe and ripe FFB, opposite to overripe FFB and loose fruits, where longer time were required.

TABLE IV: OPTIMUM STERILIZATION PROCESSING CONDITIONS FOR DIFFERENT DEGREE OF FFB RIPENESS

FFB Classification	Process Variables		Predicted Responses (DOBI)	Desirability
	X_1 (°C)	X_2 (min)	$Y_{1\text{-}4}$	
Underripe	109	20	3.612	1
Ripe	100	20	3.046	0.619
Overripe	100	69	4.462	1
Loose Fruits	100	80	2.491	0.152

IV. CONCLUSION

The present study has successfully predicted the DOBI behavior and showed promising potential for production of CPO with excellent quality of DOBI, even when different degree of FFB ripeness was used. This study however requires further works on the models verification using larger scale of sterilizer. The use of RSM has helped to identify the interactive effects between sterilization condition and CPO quality with respective optimum conditions in minimum effort and time.

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