Comparison of Response Surface Methodology and Artificial Neural Network for the Solvent Extraction of Fatty Acid Methyl Ester from Fish Waste

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Abstract

The evaluation of fatty acid methyl ester (FAME) extraction from fish waste were done by conducting experimental works using response surface methodology (RSM) and artificial neural network (ANN). The experiments were started with a preliminary experiment using one-factor-at-a-time method to evaluate the effect of temperature and mixing time on the production of FAME. Solvent extraction method was used to elucidated the best operating conditions with various temperatures (40 to 80 °C) and mixing time (2 to 6 hours) using ethanol as a solvent. The FAME profile was then analyzed using Gas Chromatography Mass Spectrometry (GCMS) after each extraction. The result showed that the mean square error (MSE) for the ANN was lower (0.026 for oil yield and 0.019 for oleic acids) compared to RSM (0.23 for oil yield and 47.16 for oleic acids). Besides, the optimization using genetic algorithm (GA) demonstrated a higher oil yield (10.65 %) and oleic acid (30.01 mg/g) than using central composite design (CCD) with 10.48 % of oil yield and 18.19 mg/g of oleic acid. Based on the MSE analysis, it revealed that ANN model produced better prediction efficiency than the RSM model. Moreover, the results showed that the effects of each factor using GA to produce oil yield and oleic acid from fish waste were accepted to be used for FAME production.

Keywords: Fish waste; solvent extraction; response surface methodology; artificial neural network; fatty acid methyl ester

Introduction

Fish waste are considered to be worthless trash and these wastes are discarded without any recovery of valuable products or nutritive compounds like protein and fatty acid. In the last few years, by-products from different types of fishes have been proposed as raw materials for the production of fish oil1. The nutrients contain in fish waste makes it high demand in market especially fatty acid such as EPA and DHA which well known as a good supplement. Fish waste is obviously can turn into something beneficial for human being and we can save world from wasting and dumping away all the fish wastes. Therefore, these different parts of fish waste have been proposed by converting the waste into fish oil.

In order to avoid wasting the fish waste and to make sure keep on supply fish oil according to high demand in market, this research is utilized fish waste to extract the fish oil. By using method that suit to demand, modified solvent extraction method is used to extract the fish oil that contain desired fatty acid from the fish waste. The extracted fish oil was analyzed by using GCMS to identify the amount of fatty acid methyl ester. Selection of the extraction parameters is important as it can produce high yield of fish oil that contain fewer impurities.

Many factors such as solvent and concentration, solvent-to-solid ratio, mixing time, temperature and pH can influence the extraction process. Thus, the optimization of the selected factors is necessary to obtain the maximum yield percentage of the desired product. Bako et al. (2018), demonstrated that Response Surface Methodology (RSM) is a technique of mathematical and statistical collection which useful for the analysis and modelling in which a response of interest is influenced by several variables to optimize the response 1. It differs from the procedure that involves test variables and changing one variable at a time. RSM examines several variables at a time, uses specific experimental designs and measures several effects to obtain the response.

Artificial neural network (ANN) is a mathematical and computational modelling technique stimulated from neural network in human brain. ANN is an important tools for modelling and map non-linear relationships between inputs and outputs disregarding of its co-relation 2. ANN has appeared as a more robust and better modelling technique as it able to clarify and create conclusions via generalization and predictive modelling of the complex non-linear process. The applications of ANN have been proven by other researcher in biological process 3. For the past few years, the genetic algorithm (GA) had been used to generate a quality solution to optimize and find problems by depending on bio-inspired operators such as crossover, selection and mutation 4. In recent times, Banerjee et al. (2016) had utilized GA method for the cultivations of *Nannochloropsis*sp. in formulated fertilizer culture medium. The study showed significant improvement in biomass and lipid productivity compared to medium optimized using response surface methodology 5.

For optimisation problems, it is common to use the mathematical and computational modelling to capture a few solutions simultaneously. The modelling tools that has been developed to solve these problems such as RSM and ANN, but all of it has its own problems. At the end, a new approach to solve the problems has been suggested. Therefore, in order to achieve the aims of this research, response surface methodology (RSM) with using central composite design (CCD) and artificial neural network (ANN) coupled with genetic algorithm (GA) approaches were conducted to evaluate the effect of temperature and time for the solvent extraction process in order to obtain the high yield of fish oil. The results obtained from RSM and ANN methods were compared to determine the best tool to optimize the extraction of fatty acid methyl ester from fish waste.

Methodology

2.1. Preparation of Fish Wastes

The raw material used for the oil extraction is fish wastes which obtained from the market in Jaya Gading Kuantan, Pahang. The fish wastes like head, viscera, fins, tail, skin, scales, and liver parts were removing and separating manually from the wastes obtained. The selected fish wastes were clean using water and dried in the oven for 24 hours at 50 °C. After that, the dried fish wastes were ground using an electrical grinder and blend it well until becomes a fine powder.

2.2. Solvent Extraction

For the extraction of fish oil, the solvent extraction was used for each extraction process. The extraction set up which comprise of thermometer, retort stand with clamp, hot plate stirrer, beaker and magnetic stirrers. Firstly, 0.72 g of fish wastes powder was weighed and mix together with 12 mL of ethanol. The test tube was added with magnetic stirrer and capped immediately to avoid vaporization using parafilm. Then, the sample was sonicated for 10 minutes to break the cell wall of the sample. After that, the test tube was clamped and put into 1 L beaker. During the extraction process, the test tube was heated in the beaker filled with water. The experiments were conducted with various temperature (40 to 80 °C) and mixing time (2 to 6 hours). After the extraction process, the sample was transferred into the centrifuge tube and centrifuge with speed of 3000 rpm for 5 minutes. After 5 minutes of centrifugation, there are oil and biomass formed in centrifuge tube at upper and bottom layer respectively. The oil that we needed was transferred into the vial using micropipette and the biomass was discharged into the bin. The vial was heated for 24 hours at 100 °C for the remaining solvent at the oil content fully evaporated. The percentage of oil yield was determined by calculating the weight of oil 6.

2.3. OFAT for Fatty Acid Methyl Ester Profile

The experiments were done to identify the highest composition (%) of fatty acid methyl ester profile by using one-factor-at-a-time (OFAT). Experiments were carried out to study the effect of two different variables which were temperature (40 to 80 °C) with 10 °C interval and mixing time (2 to 6 hours) with 1 hour interval. The highest amount of FAME was chosen for next experiments, and then the Response Surface Methodology (RSM) and Artificial Neural Network (ANN) were employed to optimize the operating parameters.

2.4. Experimental Design by Response Surface Methodology (RSM) and Optimization Method

The effect of operating parameters on the oil yield production and fatty acid composition were studied through RSM by determining the optimum conditions of mixing rate and temperature. RSM was performed through central composite design (CCD) using Design Expert Software, version 7.0.0 (State-Ease, Inc). Two independent variables were identified as production on fatty acid and oil yield. The independent variables considered in the CCD were presented in Table 1.

Table 1 - Experimental range and levels of the temperature and mixing time in the CCD

Variables	**Code**	**Levels of variables**				
		-2	**-1**	**0**	**+1**	**+2**
Temperature (°C)	A	40	50	60	70	80
Mixing Time (hr)	B	2	3	4	5	6

The experimental data were fitted by regression to a quadratic model as mention in Eq. 1:

$$Y = \beta_0 + \sum_{i=1}^{n} \beta_i X_i + \sum_{i=1}^{n} \beta_{ii} X_i^2 + \sum_{i=1}^{n} . \sum_{j=1}^{n} \beta_{ij} X_i X_j + \epsilon \quad (1)$$

where Y represents the value of the predicted response $\beta_0$is a constant, β_i, β_{ii}, and $\beta_{ij}$are the linear, quadratic and interaction coefficients, respectively, and $X_i$and $X_j$are the experimental variables which levels are being optimized. Thirteen run were performed (Table 2) and the responses were analyzed using Analysis of Variance (ANOVA) for the production of oil yield (%) and FAME (mg/g). Based on the result of FAME in the previous preliminary experiment in Sec 2.3, oleic acid gives the highest concentration compared to palmitic acid, palmitoleic acid, stearic acid and linoleic acid. Therefore, oleic acid was chosen as Response 2 in RSM.

2.5. Validation Experiment

Maximum oil yield and oleic acid were predicted using the optimum conditions where the temperature and mixing time were set at 70 °C and 3.28 hours, respectively. A validation experiment was performed in triplicates under the optimized conditions and the results were compared with predicted value to confirm the validity of the model.

2.6. ArtificialNetwork Modelling and Genetic Algorithm Optimization

The model had used basic architecture of 2 input node for temperature and mixing time and one output node for the oil yield percentage or one output for oleic acids weight. Two models were generated for each output to follow the regression modelling. Only the output node used linear activation function while the hidden node used hyperbolic activation function. The implementation of the ANN modelling had used the ENCOG 3.3.0, Java library with Netbeans 8.0.2 was used as the development IDE.

Table 2 - Experimental design and measured values for oil yield and oleic acid

Standard Order	**Coded values of variables**		**Oil Yield (%)**	**Oleic Acid (mg/g)**
	A	**B**		
1	50	3	9.86	25.55
2	70	3	10.42	24.02
3	50	5	10.00	25.1
4	70	5	9.58	18.71
5	40	4	9.72	26.85
6	80	4	10.56	24.19
7	60	2	10.14	6.10
8	60	6	9.31	19.46
9	60	4	10.28	24.35
10	60	4	9.86	25.93
11	60	4	10.14	25.15
12	60	4	10.00	22.33
13	60	4	9.86	28.47

The ratio of dataset was 80:20 with 10 random samples of dataset training and 3 samples testing set was used in this modelling. The dataset was normalized between -1 to 1 and the network performance will be measured using mean square error (MSE) for comparison with the polynomial regression model (RSM).The optimization of the neural network output was implemented using Jenetics 3.6.0. The population size was set to 30 chromosomes with 100 generations for all the runs.

2.7. Fatty Acid Methyl Ester Analysis

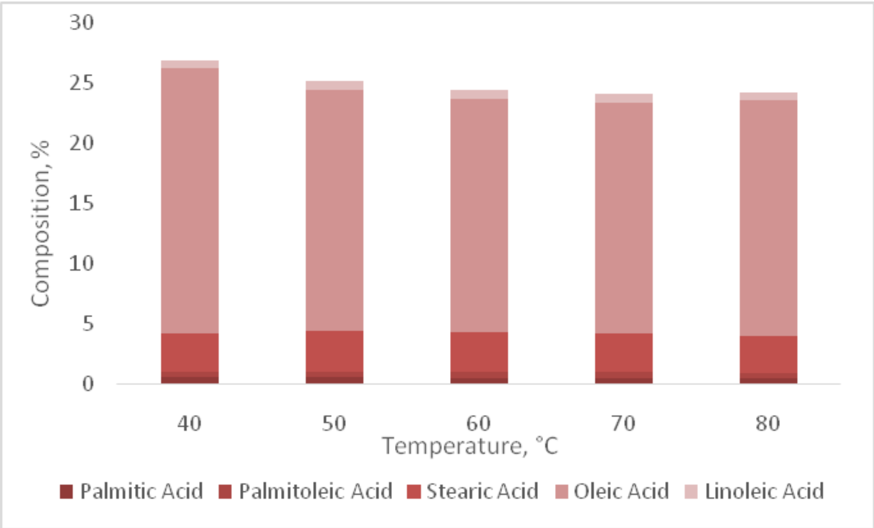
The fatty acid profiles in the extracted fish oil was analyzed by using Gas Chromatography Mass Spectrometry (GCMS) Agilent Technologies (G3171A, China) after each extraction process 7. 1 μL of extracted fish oil samples for each parameter at optimum condition was injected into GCMS. The compound of fatty acid methyl ester were identified with the standard of C16:0, C16:1, C18:0, C18:1, C18:2 and C18:3. Details of the analysis procedure have been provided elsewhere 8.

3. Results and Discussion

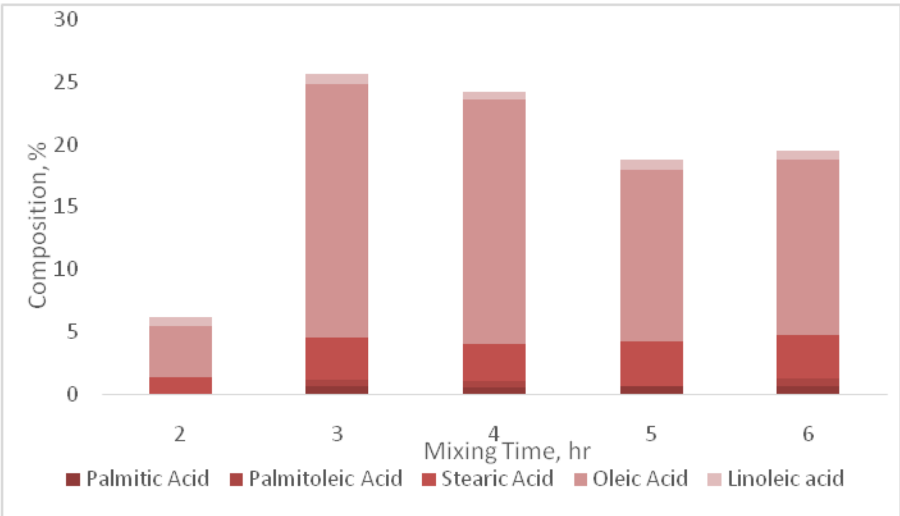
3.1. Fatty Acid Methyl Ester (FAME) Profile of Fish Oil

In this current study, a preliminary study of fish oil was extracted from fish wastes by the solvent extraction method using the method of OFAT. The amount of FAME (%) was identified from the chemical analysis of GCMS. The FAME obtained from fish waste was analyzed at different temperature (40, 50, 60, 70 and 80 °C) and mixing time (2, 3, 4, 5 and 6 hours).

Fig. 1 shows the composition of FAME (%) which affected by temperature and mixing time. There are five FAME compounds detected in the fish oil which are oleic acid, palmitic acid, palmitoleic acid, stearic acid and linoleic acid. Based on the figure, oleic acid shows the highest percentage of FAME compositionscompared to other FAME after extraction of fish oil from fish waste. However, there are only small amount of palmitic acid, palmitoleic acid and linoleic acid identified in fish oil at all range of temperature and mixing time. Based on Fig. 1(a), among of five temperatures used, the highest composition of oleic acid obtained at the temperature of 40 °C which is 21.89%. It is also shows that the percentage of oleic acid decreased as the temperature increase until 70 °C and keep maintain the trend even the temperature increase. Meanwhile, Fig. 1(b) shows the different pattern for the composition of oleic acid where the highest percentage obtained at 3 hours of mixing time.



(a)



(b)

Fig. 1 - The effect of (a) temperature and (b) mixing time on the composition of FAME

The major constitutes of FAME was consistently of oleic acid. The presence of oleic acids was in agreement with Khoddami et al. (2009) who claimed that oleic acid (C18:1) act as major components in fish oil 9. The ratio of oleic to linoleic acid was considered an important criterion to evaluate the quality of oil. Increasing the ratio by increasing oleic acid and decreasing linoleic acid deliberates better stability and longer shelf life.

Fatty acid profiles of the oil extracted from the fish waste for two different samples are shown in Table 3. Sample 1 represents the effect of temperature at 40 °C while Sample 2 was taken from the effect of mixing time at 3 hours. These samples were chosen based on the highest amount of oleic acid obtained in this research. GCMS analysis illustrates that the sample were containing of the common fatty acid methyl ester which were consisted of oleic acid (C18:1), stearic acid (C18:0), palmitic acid (C16:0), palmitoleic acid (C16:1) and linoleic acid (C18:2).

With reference to Table 3, the fatty acids of the fish oil were generally composed of saturated fatty acid, SFA (palmitic acid and stearic acid) and unsaturated fatty acid, UFA (palmitoleic acid, oleic acid and linoleic acid). The extracted fish oil for both samples contained greater percentage of UFA which is 23.08% and 21.56%, respectively. While the amount of SFA obtained were much lower than UFA which are 3.77% and 3.99% for sample 1 and sample 2, respectively. The SFA in both sample were palmitic acid and stearic acid with the highest level of C16:0 (0.60%) and C18:0 (3.39%) were determined in sample 2. The UFA identified was predominantly compose of oleic acid (C18:1) followed by linoleic acid (C18:2) and palmitoleic acid (C16:1). The highest oleic acid were obtained in sample 1 (21.89%), compared to the sample 2 (20.34%). In comparison between Sample 1 and 2, the amount of each FAME is not much different for both samples.

Table 3 - Fatty acid profile of fish wastes

FAME	**Amount of FAME (%)**	
	Sample 1	**Sample 2**
C16:0	0.60	0.60
C16:1	0.50	0.49
C18:0	3.17	3.39
C18:1	21.89	20.34
C18:2	0.69	0.73
SFA	3.77	3.99
UFA	23.08	21.56
Total FAME	26.85	25.55

FAME of fish oils were found suitable for use as biodiesel in diesel engine. The most important biodiesel properties are carbon number, oxidative stability and energy content. The fatty acid chain with more highly unsaturated (which is a double bond of the carbon to carbon bonds), will produce lower gel point. Normally, biodiesel made from highly UFA will require an oxidative stabilizer to be used safely as fuel. The heating value of a fuel also increases with increasing carbon number in fuel molecules. Therefore, UFA is identified as excellent FAME for cold weather biodiesel production and use. In this case, UFA which is oleic acid (C18:1) was chosen as the highest percentage composition and the longest carbon among others, so oleic acid was chosen as response for next study which are response surface methodology (RSM) and artificial neural network (ANN).

3.2. Response Surface Methodology (RSM)

3.2.1. Regression Model and Statistical Analysis. The second order polynomial model was utilized the relationships between the response (oil yield and oleic acid) and two affected factors (mixing time and temperature) which could give the predicted value of the following response:

$$\text{Oil yield} = 10.02 + 0.15\,A - 0.2\,B - 0.25\,AB + 0.028\,A^2 - 0.076\,B^2 \quad (3)$$

$$\text{Oleic acid} = 20.51 + 1.00\,A + 1.20\,B - 1.22\,AB + 0.11\,A^2 - 2.80\,B^2 \quad (4)$$

where the oil yield and oleic acid were the response, and A and B represent the temperature and mixing time, respectively. The term of A and B are denoted as the main effects, while AB was the interaction involved between the factors. Quadratic effects were presented through A^2 and B^2 to presence of curvature in the model.

3.2.2. Analysis of Variance (ANOVA). For more detailed explanation of mean squares, F values and p-values, ANOVA was applied by fitting the experimental data in Table 4 and 5. According to the results, p-value for both result were 0.0098 and 0.0301, respectively. If p-value is less than 0.05, it showed that the model terms were significant 10. In this study, both of the models show A^2 and B^2 are significant model terms. Meanwhile, the "lack of fit" was not significant with p-value which is 0.5339 and 0.1557 for both of the models, signified that the model adequately explained the data in the region of experimentation. The lack of fit test was used to check the adequacy of the model 11. By referring to the results in Table 4 and 5, the coefficient of determination, R^2 values obtained were 0.8431 and 0.8781 in which it demonstrated that the model was well fitted for the predicted and experimental data.

3.3. Artificial Neural Network (ANN) Modelling

The generated model for oil yield modelling was obtained from the network structure of 2-5-3-1-1 with its model MSE for 10 runs is shown in Table 6. All the runs showed similar low training and testing MSE to all the models thus the training process successfully produced low error networks. The resultwas summarized in Fig. 2. The model run number 2 was selected as the best model and will be used in optimization process because it had the lowest testing MSE while all runs have same training MSE of 0.010036.

Table 4 - ANOVA analysis for oil yield

Source	**Sum of Squares**	**Mean Square**	**F Value**	***p*-value Prob > F**	
Model	1.17	0.23	7.52	0.0098	Significant
A-Temperature	0.28	0.28	8.85	0.0207	
B-Mixing Time	0.46	0.46	14.88	0.0062	
AB	0.24	0.24	7.70	0.0275	
A^2	0.018	0.018	0.57	0.4768	
B^2	0.13	0.13	4.24	0.0784	
Residual	0.22	0.031			
Lack of Fit	0.085	0.028	0.85	0.5339	not significant
Pure Error	0.13	0.033			
Cor Total	1.39				
R-Squared	0.8431				
Adj R-Squared	0.7310				

Table 5 - ANOVA analysis for oleic acids

Source	Sum of Squares	Mean Square	F Value	*p*-value Prob > F	
Model	235.79	47.16	4.91	0.0301	Significant
A-Temperature	12.06	12.06	1.26	0.2994	
B-Mixing Time	17.30	17.30	1.80	0.2214	
AB	5.98	5.98	0.62	0.4560	
A^2	0.29	0.29	0.031	0.8660	
B^2	179.86	179.86	18.73	0.0034	
Residual	67.23	9.60			
Lack of Fit	46.71	15.57	3.04	0.1557	not significant
Pure Error	20.52	5.13			
Cor Total	303.02				
R-Squared	0.8781				
Adj R-Squared	0.6197				

Table 6 - ANOVA analysis for oleic acids

Model	Training MSE	Testing MSE
Run 1	0.010036	0.083561
Run 2	0.010036	0.083466
Run 3	0.010036	0.083524
Run 4	0.010036	0.083571
Run 5	0.010036	0.083572
Run 6	0.010036	0.083541
Run 7	0.010036	0.083567
Run 8	0.010036	0.083495
Run 9	0.010036	0.08359
Run 10	0.010036	0.083698

The result for oleic acids modelling training and testing MSE is shown in Table 7. Network architecture of 2-3-1-1 was the architecture which produced significant models with error of 0.01. The training MSE were varied from the 0.01 to lowest of 0.0096 but still within targeted MSE. Fig. 3 shows the graph which compared the training and testing MSE. The best model was run number 1 because it also has the lowest testing MSE among all the models. A small value of MSE indicates that the model had a better prediction efficiency 12. This model will be used to optimize the oleic acids output.

Table 7 - ANOVA analysis for oleic acids

Model	Training MSE	Testing MSE
Run 1	0.009992	0.052193
Run 2	0.010005	0.054957
Run 3	0.009967	0.061911
Run 4	0.009987	0.053502
Run 5	0.009983	0.062961
Run 6	0.009609	0.055193
Run 7	0.009983	0.06067
Run 8	0.00985	0.058892
Run 9	0.009985	0.058098
Run 10	0.009974	0.053379

The ANN modelling comparison with the RSM model is shown in Table 8 using the best selected respective ANN model above. The overall MSE were used for this comparison where both training and testing MSE was combined for overall model MSE. The ANN modelling showed very low MSE compared both the regression models thus the ANN model had better performance to model the processes. This discovery was similar with the study conducted by Ciric et al. (2020) that concluded that ANN model produced a better prediction and estimation capabilities 2. Furthermore, the comparison of mathematical modelling between artificial intelligence (DE) and RSM was made by Jahromi et al. (2018) where the methods are developed for the optimization in ethylene plant. The result shows that DE method provide the profit percentage of 61.6% more than RSM method and it shows the ability of DE to produce more profit in chemical plant in comparison with RSM 13.

Table 8 - ANOVA analysis for oleic acids

Model	Response	MSE
RSM	Oil yield	0.23
	Oleic acids	47.16
ANN	Oil yield	0.026
	Oleic acids	0.019

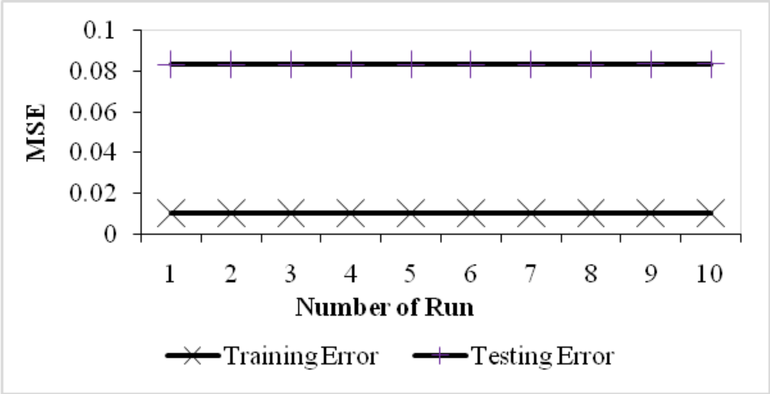


Fig. 2 - Oil yield modelling result

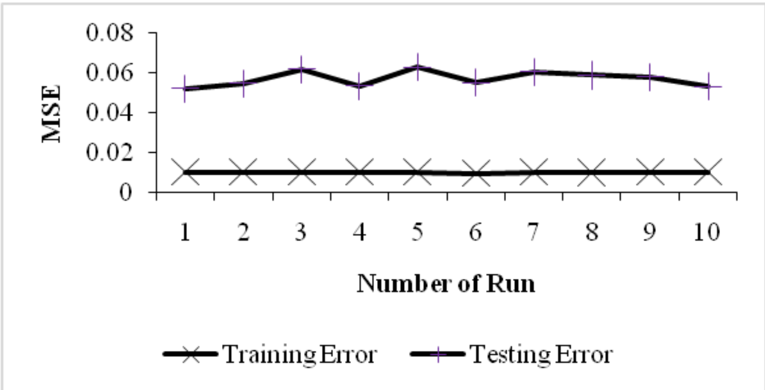
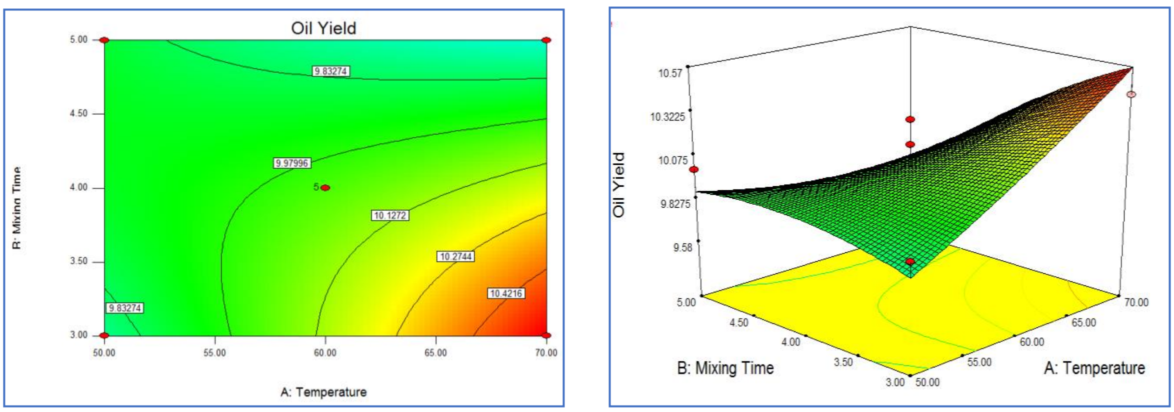


Fig. 3 - Oleic acids modelling result

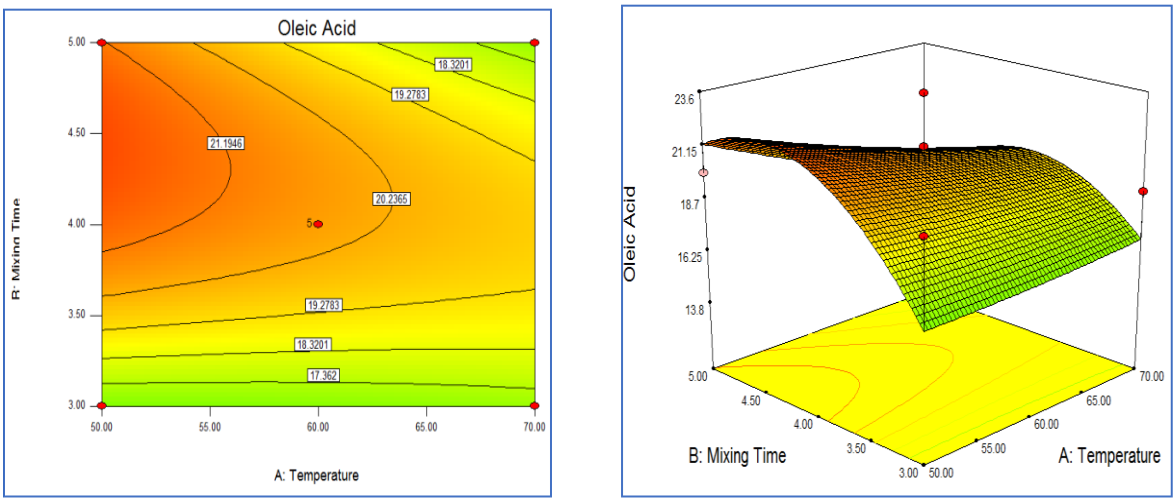
3.4. Optimization Results using Central Composite Design (CCD)

The response surface plots of interaction between temperature and mixing time are shown in Fig 4 and 5. Fig 4 demonstrates the main relationship between temperature and mixing time for the oil yield production for 2D contour plot and 3D view of shaped curve. The oil yield production increased when the temperature increased and reached the maximum oil yield of 10.57 % which above the predicted value (10.48 %). Meanwhile, Fig. 4 presents the correlation of two factors for the production of oleic acid. In this case, the oleic acid just had a slightly difference value between high temperature (70 °C) and low temperature (50 °C), but the result of oleic acids obtained still below the predicted value of 18.19 mg/g.



(a) **(b)**

Fig. 4 - Response surface plot (a) 2D contour plot (b) 3D view for the effect of interaction between temperature and mixing time on production of oil yield



(a) (b)

Fig. 5 - Response surface plot (a) 2D contour plot (b) 3D view for the effect of interaction between temperature and mixing time on production of oleic acid

3.4.1 Validations of the Model. The predicted value to select the highest oil yield and oleic acid is given in Table 9. Validation experiment was run in order to validate the predicted value. The maximum response for oil yield and oleic acid obtained from the experiment were 18.19 % and 23.12 mg/g, respectively, whereas the predicted value using Eq. 3 and 4 is 10.48 % and 18.19 mg/g, respectively. The error for oil yield and oleic acid is very high which is 76.24 % and 36.89 %, respectively. This might be happened because of the fish waste stored for too long, so the waste sample exposed to the surrounding and the environmental effect will damage the sample which were affected the result.

Table 9 - Validation result of optimum conditions from CCD

Run	Oil Yield (%)			Oleic Acid (mg/g)		
	Predicted Value	Experimental Value	Error Percentage	Predicted Value	Experimental Value	Error Percentage
1	10.48	18.19	73.57	18.19	11.48	36.89
2	10.48	18.33	74.90	18.19	11.52	36.67
3	10.48	18.47	76.24	18.19	11.66	35.90

3.5. Optimization Result using Genetic Algorithm (GA)

The oil yield output optimization was done using five GA optimization runs which used the ANN model number 2 as the fitness function. Table 10 shows the oil yield optimization and the run number have the highest oil yield from the model output maximization. The result shows the normalize value of the oil yield and the denormalized value of oil yield in percentage. It gave higher value than the predicted value by CCD but lower than the experimental value. Optimization run number 3 produced the highest oil yield with 10.65 %.

Output optimization of the oleic acids is shown in Table 11. The GA optimization produced much higher oleic acids than the predicted and experimental value using RSM with the highest oleic acids was 30.0139 mg/g. Thus, it shows a higher result was obtained by using GA and GA is a suitable tool for optimization of fatty acid methyl ester extracted from fish waste. This result was in line with Banerjee et al. (2016) in which the study demonstrated a significant improvement in biomass and lipid productivity by using GA compared to medium optimized using response surface methodology 5. Jahromi et al. (2018) have also presented a comparative study between differential evolution algorithms (DEA) and Box-Behnken method (BBD). This algorithm is different with genetic algorithms studied in this current research because the DEA is based on the principle of evaluation theory 13. However, both algorithms (DEA and GA) are trying to approach the optimal results with a random but the ability of algorithms to find the optimum solutions is still in intelligently search.

Table 10 - Oil yield optimization using GA

Optimization	Temperature	Mixing time	Oil yield normalized	Oil yield (%)
Run 1	0.954838	-0.92142	1.14146	10.64841
Run 2	0.978221	-0.97009	1.141598	10.6485
Run 3	0.995121	-0.97763	1.141646	10.64853
Run 4	0.885366	-0.92375	1.141136	10.64821
Run 5	0.95718	-0.86769	1.141368	10.64835

Table 11 - Oleic acids optimization using GA

Optimization	Temperature	Mixing time	Oleic acids normalized	Oleic acids (mg/g)
Run 1	-0.9948	-0.96484	1.139976	30.01394
Run 2	-0.93018	-0.86524	1.139631	30.01013
Run 3	-0.88179	-0.96645	1.139895	30.01305
Run 4	-0.8953	-0.9906	1.139966	30.01382
Run 5	-0.72607	-0.98562	1.139783	30.0118

4. Conclusion

A correlation analysis between classical one-factor-at-a-time (OFAT) approach and statistical experimental design using a response surface methodology (RSM) model with central composite design (CCD) and artificial neural network (ANN) coupled with genetic algorithm (GA) for optimization were done in this research. The result showed that the model from ANN produced a better prediction with lower mean square error (MSE)

(0.026 for oil yield and 0.016 for oleic acids) than by using RSM (0.23 for oil yield and 47.16 for oleic acids). Furthermore, the optimization using genetic algorithm (GA) verified a higher oil yield (10.65 %) and oleic acid (30.01 mg/g) than using RSM (10.48 % oil yield and 18.19 mg/g). This study revealed that ANN model produced better prediction efficiency than the RSM model. Moreover, the results showed that the effects of each factor using GA have a significant influenced on the oil yield and oleic acid production for biodiesel production in the future

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