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Study of the Lipid Oxidation for Shelf – Life Prediction of Mayonnaise using Arrhenius Equation

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Abstract:
In this study, the shelf life of mayonnaise (product of high lipid content) was rapidly predicted by studying the lipid oxidation reaction. Samples of mayonnaise were stored in glass bottles and stored in incubators at 30ºC, 37ºC and 45ºC for 2 months to follow the perishable degree of product by measuring the acid and peroxide values every 7 days. Results showedthat the lipid hydrolysis follows zero-order reaction mechanism with activation energy Ea = 33.00 kJ / mol, the acid value increase slowly within study. The Arrhenius equationwas establishedaccordingto theperoxide value: y = -4054.7x + 5.5563 (R^2 = 1), estimated shelf life was13.7 months.Lipid oxidation followed the first-order reaction mechanism with the activation energy E_a = 24.76 kJ/mol, the peroxide value increased significantly within study. The Arrhenius equationwas establishedaccordingto theperoxide value: y = -2977.6x + 6.4962 (R^2 = 0.9555), the predicted shelf life of the sample was 6,0 months, matches with the published shelf life given by the manufacturer. Thus, lipid oxidation reaction is the main damaging factor of this product and hence, we can confirm the accuracy and applicability of mathematical modeling method to quickly predict the shelf life of food products.

Keywords: *lipid oxidation, acid value, peroxide value, Arrhenius equation, mayonnaise sauce, predict shelf life; accelerated testing*

I. INTRODUCTION

For a food product, shelf life is an important parameter Shelf life is the period in which food products are required to be safe and sustainable under regulated conditions during storage, distribution, retail and use [1]. After that time, the product is not allowed to be available in the market [2]. Precise evaluation relating to sheft life can determine both the quality of products and thei popularity after releasing [2]. On the other hand, while food companies have always carried on researches for new products. It is noticed that the more quickly the shelf life is determined, the easier these products can be prevailed in the market.

Direct observation of products in the same storage conditions on the market is the most accurate method of determining the shelf life [1]. However, by using this method, the product must be stored under recommended storage conditions, monitored and checked until the product is damaged, then based on recorded data to establish the shelf life. Therefore, the execution time will be long (especially for products that are difficult to damage) and the product will not be consumed soon.

Following methods to predict quickly the shelf life of food products are deduced:

- The method based on the shelf life of similar products, this method has various risks because only a slight difference in materials or manufacturing can change the shelf life [1], [3].
- Thermal acceleration method (Q10): use temperature higher than normal storage temperature to accelerate the deterioration (aging) of the product, then monitor and check until the product is damaged completely by using the sensory evaluation method [1].
- Mathematical modeling method: products stored in harsh conditions which are similar to those in Q10 but only suitable analytical criteria are selected for evaluation (chemical and physical criteria,

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microorganisms, etc.) [1]. Determining the shelf life of the food product by using this method gives quick results without exposuring until the product is completely damaged, and no sensory evaluation is required.

Therefore, this method has many advantages such as time - saving, money – saving and has been widely studied by many scientists in the world. Meanwhile, in Vietnam, there haven't been any researches adopting mathematical modeling method for the quick prediction of the shelf life in the food industry.

Mayonnaise is a condiment sauce obtained by emulsifying edible oil(s) in a aqueous phase consisting of vinegar, the oil-in-water emulsion being produced by egg yolk [4]. This product made from a variety of ingredients with very different properties, so it is difficult to determine the shelf life of it [2]. In fact, the shelf life is still determined by Q10 method. In addition, manufacturers constantly innovate mayonnaise product formulas, while only a slight change in the ingredients or manufacturing process can change the shelf life of the product [2].

Therefore, we have conducted on the study: "Study of the lipid oxidation for shelf – life prediction of mayonnaise using Arrhenius equation" to investigate the accuracy and applicability of this method in practice to help producers save time and costs. At the same time, the study aims to quickly determine the shelf life of other food products.

II. STUDY OBJECTS AND METHODS

2.1. Materials method of saving analysis samples

The study object used in this study is the Aji - Mayo Mayonnaise Sauce of The Ajinomoto Vietnam Co., Ltd., that contains the following ingredients: vegetable oil, water, fresh egg, egg powder, sugar, salt, vinegar, malt, flavour enhancer (E621), acid modifier (E260, E330), stabilizer (E1442, E415), color (E160a(1)), preservative (E202, E386) and antioxidant (E321) listed on the brand label, the manufacturer's published shelf-life is 6 months. Products used for research were purchased at supermarkets in Da Nang city, Vietnam and they are products whoseroduction date was closest to the sampling date (Figure 1).

The production date of the product was 05.02.2018, while the sampling date was 05.03. 2018. All products used for research were in the same batch.

The research of Elmlund and his colleagues showed that at high temperatures above 50ºC, food products may have many mutations which can not observe during product storage at normal temperature condition [5].

Therefore, mayonnaise in the bottles was well - mixed homogeneous, then put into 100 ml capacity clear glass vials, 100g mayonnaise per vial, then stored in the incubator at 3 temperatures of 30ºC, 37ºC, 45ºC (Figure 2). Samples were stored in incubators for 7 weeks. Each week, from each incubator, took off 1 sample vial for analysis, measured acid value, peroxide value of samples at the 0th, 7th, 14th, 21th, 28th, 35th, 42th and 49th day, respectively

Figure 1. *Mayonnaise Aji-Mayo*

Figure 2.*Mayonnaise samples are stored in incubator*

2.2. Accelerated shelf life testing of mayonnaise by using mathematical modeling method

To quickly determine the shelf life, a quality factor of the product is selected to test and evaluate it's variation over time[6], [7]. The change of quality factor A of the product is calculated according to the following equation:

$\frac{dA}{dt} = k[A]^n$ or F(A) = kt

In which: k is the kinetic constant, [A] is the concentration of the quality factor A, n is order of reaction, F(A) is the change of quality factor A that depends heavily on the order reaction n [5].

Free fatty acids (FFA) are produced from triacylglycerides (TAG) through chemical or enzymatic hydrolysis. They are usually associated with undesirable flavour and textural changes when they are present in fats and oils [8]. Mayonnaise is the rich fatty product, so the deterioration of this reflected by the hydrolysis reaction (the acid value of fat) and the oxidation reaction (the peroxide value of fat) is chosen as the main damaging factor. And temperature is the factor used to speed up the deterioration of the product to quickly determine the shelf life.

Arrhenius model is used to represent the relationship between the rate of hydrolysis reaction and oxidation reaction and temperature change [7], [9]. The Arrhenius equation is represented by:

$k = k_0 \, e^{-\frac{E_a}{RT}}$ or $lnk = lnk_0 - \frac{E_a}{R}\left(\frac{1}{T}\right)$

In which: k_0 is the Arrhenius equation constant, E_a: is the energy of activation (kJ/mol), T: is the absolute temperature (product storage temperature), R: is the gas constant, R = 8,3144 (J/mol K) [1], [10].

Determine the shelf life of a food product, following 3 steps:

- Determine the reaction rate constant k at each study temperature: plot the following equations [A] – t, ln (A) – t, 1/[A] – t. Select the order of reaction corresponding to the equation with the largest R^2. In that case, the reaction rate constant k at each temperature is equal to the slope of the plot at the corresponding temperature [5].

- Determine the reaction rate k_T at the actual storage temperature by plotting the Arrhenius equation $\ln k = \ln k_0 - \frac{E_a}{R}(\frac{1}{T})$. Replace the T value in the equation with the actual storage temperature; we get k_T value - the reaction rate at the actual storage temperature.

- Calculate shelf life at actual storage temperature T according to the formula:

+ t = $\frac{A_1 - A_0}{k_T}$ if reaction order is 0,

+ t = $\frac{\ln(A_1) - \ln(A_0)}{k_T}$ if reaction order is 1.

In which: A_1, A_0 are respectively the critical concentration and the initial concentration of the quality factor A [1], [10], [11].

Result evaluation: compare the shelf life calculated in this method with the published expiry date of the product to comment on the accuracy and applicability of this method.

2.3. Method for fat extraction and measuring the axit and peroxide value of fat in the study sample

To accurately determine the acid value and the peroxide value, the fat (lipid) needs to be separated from the emulsion system by a moderate method (do not use heat) to avoid undesirable effects on these indicators.

In this study, we used the methodology, written on the GOST 31762 – 2012: Mayonnaise and mayonnaise sauce. Acceptance rules and testing methods, Russian [12] to extract lipid from mayonnaise. Follow that, samples are taken off from the incubator, thoroughly mixed to reach homogeous state. Then take about 10 ÷ 15 g simple to put into a 250 ml erlenmeyer flask, add gradually 10 ÷ 15 ml of alcohol to the that flask, combine with stirring gently, the emulsion system will be gradually destroyed, then add 20 ÷ 30 ml of chloroform slowly into the flask, stirring gently. Shake well about 30 minutes to increase fat separation effect. After that, the entire solution in the flask is transferred to a falcon tube, centrifuged at a speed of 3000 rpm within 3 minutes. After centrifugation, the solution in the falcon tube will be divided into 2 layers, using

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a pipette to remove the upper layer. The rest is filtered through a sheet of filter paper wetted with chloroform, then wash residue on the filter paper with maximum 10 ml of chloroform to avoid loss of fat in the residue [12].

The acid value of the fat is titrated with KOH 0,1 N with a few drops of phenolphthalein until a persistent pink color appears in 1 minute [12].

The peroxide value of the fat was titrated with sodium thiosulfate $Na_2S_2O_3$ 0.001 N until the solution losts color in 30 seconds with a few drops of starch adhesive [10]. The peroxide value, expressed in mmol of active oxygen per 1 kg of fat was calculated according to the folowing formula:

$$PV = \frac{(V_2 - V_1) * C_M * 1000}{m}$$

where, V_1: the volume of $Na_2S_2O_3$ solution used for fat simple titration (ml); V_2: the volume of $Na_2S_2O_3$ solution used for blank simple titration (ml); C_M: $Na_2S_2O_3$ (0,05 M) solution concentration; m: fat simple mass (g).

2.4. Methods for determining the chemical compositions of the study sample and statistical analysis

Determine the moisture content, the lipid content, the total amount of ash and the protein content by the methods according to OAOC 1990[13].

All treatments in this study were repeated 3 times. Results were calculated on Excel and one-way ANOVA software, the significance of the results were applied with 95% of confidene.

III. Result and Discussion

3.1. Chemical compositions of the study sample

Some of the main chemical components of mayonnaise are shown in Table 1. From Table 1, we can see that mayonnaise is a rich fatty product (57.52 %) and low moisture content (19.28 %). Oxidative deterioration is a major quality loss in foods. In addition, several intrinsic and extrinsic factors, including the content and composition of unsaturated fatty acids, the storage temperature and the concentration and activity of antioxidant substances can influence the oxidative status of food [14], [15]. During the storage, the lipid oxidation is faster than the protein oxidation because the primary (hydroperoxides) and secondary lipid oxidation products (aldehydes and ketones) can react with proteins and further induce protein oxidation [15]. It is a characteristic form of the deterioration, we can be based on variation of lipid to predict shelf life of mayonnaise.

Table - 1 Some of the main chemical components of mayonnaise

No.	Component	Content	unit
1	Moisture	19.28 ± 0.01	% raw material
2	Ash	2.80 ± 0.01	% dry substance
3	Protein	1.09 ± 0.04	% dry substance
4	Lipid	57.52 ± 0.02	% raw material

3.2. Determine the shelf life of mayonnaise through the determination of acid value (AV)

The results of determining the acid value of the product after 49 days at 3 different temperatures are shown in Table 2. The results in Table 2 show that the acid value of samples increases significantly slowly with the storage time. The reason is that in lipid hydrolysis, water content plays an important role by affecting interfacial area [16]: mayonnaise has a low moisture content, therefore, hydrolysis reaction occurs slowly, the acid value does not increase much during the sample storage time.

Beside that, the acid value increases with the rising of the storage temperature: After 49 days the acid value of mayonnaise increases 31 % at 30° C (303K), 45.5 % at 37° C (310K) and 65.78 % at 40° C (318K), respectively, in comparing with 0 day storage. As N.A. Serri reported that temperature may affect the oil hydrolysis reaction in a positive way or vice versa: A rise in temperature will increase the reaction rate as explained by the transition state theory [17].

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Table - 2. Acid value of mayonnaise samples in 49 days at different storage temperatures

Day	303 K	310 K	318 K
0	**0.0637 ± 0.0004[Aa*]**	**0.0637 ± 0.0004[Aa]**	**0.0637 ± 0.0004[Aa]**
7	**0.0671 ± 0.0017[Aab]**	**0.0706 ± 0.0005[Ab]**	**0.0725 ± 0.0003[Bb]**
14	**0.0702 ± 0.0007[Ab]**	**0.0720 ± 0.0028[Bbc]**	**0.0761 ± 0.0019[Bc]**
21	**0.0749 ± 0.0014[Ac]**	**0.0766 ± 0.0007[Bcd]**	**0.0833 ± 0.0007[Bd]**
28	**0.0761 ± 0.0017[Acd]**	**0.0833 ± 0.0026[Bd]**	**0.0923 ± 0.0007[Ce]**
35	**0.0796 ± 0.0017[Ade]**	**0.0864 ± 0.0009[Be]**	**0.0960 ± 0.0010[Cf]**
42	**0.0803 ± 0.0028[Ae]**	**0.0870 ± 0.0007[Be]**	**0.1001 ± 0.0012[Cg]**
49	**0.0835 ± 0.0010[Ae]**	**0.0927 ± 0.0003[Bf]**	**0.1056 ± 0.0013[Ch]**

** The different letters in the table show a significant difference between the values when analyzing by one-way ANOVA with p<0.05*

Plot the equation AV-t, lnAV-t, 1/AV-t (Figure 3, 4, 5), compare relevant determination coefficients (R^2) to determine the order reaction. The R^2 coefficients of the plot in order reaction 0 is the largest (Figure 3), so the hydrolysis reaction of fat is zero-order reaction, it means that the reaction rate is independent of the concentration of reactant substances. Since the reaction order is 0, the reaction rate constant at each temperature is the slope of the plot AV-t corresponding to that temperature (Table 3).

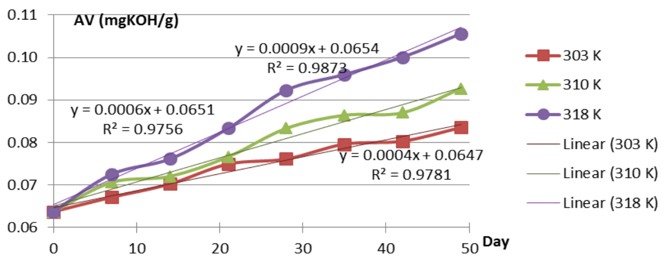

Figure 3. The change of acid value over time (AV – t)

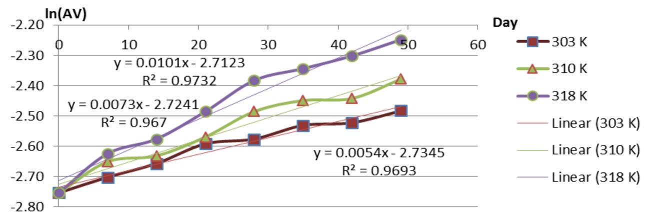

Figure 4. The change of ln(AV) over time (lnAV – t)

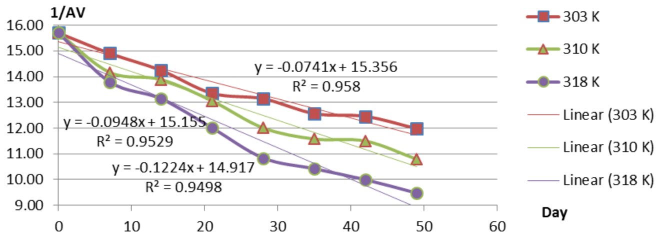

Figure 5. The change of 1/AV over time (1/AV – t)

Table -3 The reaction rate constant corresponds to the temperatures

T (K)	k	1/T	ln(k)
303	**0.0017**	**0.0033**	**-6.377**
310	**0.0025**	**0.0032**	**-5.991**
318	**0.0037**	**0.0031**	**-5.599**

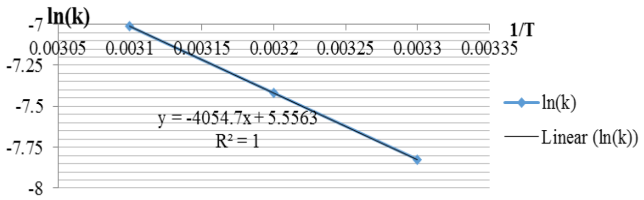

Figure 6. Arrhenius eqution plot

The equation lnk = a$\frac{1}{T}$ + b forms lnk = -4054.7$\frac{1}{T}$ + 5.5563, R^2 = 1 (Figure 6). The kinetic parameters for the lipid hydrolysys rate of mayonnaise include: The energy of activation: Ea = 33.71 KJ/mol; Arrhenius equation constant: k_o = 258.86.

Suppose the actual product storage temperature is 30ºC (303K), lnk = $-4054.7\frac{1}{303} + 5.5563$. Inferred k = 0.0017.

According to Russian Customs Union Technical Regulation on fat and oil products, the critical value (A_1) of acidvalue of mayonnaise is 0.6 mg KOH/g [18]. The initial acidvalue (A_0) of the sample is 0.0637 mg KOH/g.

Shelf life of products at 30ºC (303 K) is calculated according to formula:

$$t = \frac{A_1 - A_0}{k} = \frac{0,6 - 0.0637}{0.0017} = 383 \text{ (days)}$$

The sample has started to be analysed at the 28th day since products was produced. So, according to the results of acid value study, the shelf life of the product if stored at 30ºC is: 383 + 28 = 411 days ≈ 13.7 months, inconsistent with the manufacturer's published (6 months).

In other words, it is not possible to build a mathematical modeling to quickly predict the shelf life of mayonnaise products by monitoring acid value. It means that lipid hydrolysis reaction is not the main damaging factor of this product.

3.3. Determine the shelf life of mayonnaise through the determination of peroxide value (PV)

The results of determining the peroxide value of the product after 7 weeks at 3 different temperatures are shown in Table 4.

The results in Table 4 show that, compared to the very slow increase of acid value, peroxide value of mayonnaise increases very rapidly with the time storage ($p<0.05$). After 49 days the peroxide value of mayonnaise increases 409 % at 30° C (303K), 919.5 % at 37° C (310K) and 1502.43 % at 40° C (318K), respectively, in comparing with 0 day storage.

Futhermore, the temperature storage shows the signicant effect on the peroxide value. At the 49th day storage the PV of mayonnaise stored at 37° C is higher than 2 times and at 40° C – 3.15 times, re respectively, than the PV of mayonnaise stored at 30^0C.

Thus, the change in the peroxide value of the sample depends heavily on the temperature, the higher the temperature, the faster the rate of change. The peroxide value of the product changing over time is regarded as result of lipid oxidation reaction. This reaction occurs through several stages, can happen by lipid itself, without the participation of other substances and is heavily affected by temperature.

These results were similar lipid oxidation behavior reported by the other authors who worked with coconut oil [19], olive oil [20], vegetable oil [21] or butter [22].

Table - 4 Peroxide value of mayonnaise samples in 49 days at different storage temperatures

Day	**303 K**	**310 K**	**318 K**
0	**0.041±0.0035^{Aa*}**	**0.041±0.0035^{Aa}**	**0.041±0.004^{Aa}**
7	**0.057±0.0058^{Ab}**	**0.066±0.0063^{Bb}**	**0.082±0.007^{Bb}**
14	**0.07±0.0007^{Ac}**	**0.105±0.0066^{Bc}**	**0.141±0.001^{Cc}**
21	**0.09±0.0035^{Ad}**	**0.151±0.0080^{Bd}**	**0.205±0.007^{Cd}**
28	**0.12±0.0023^{Ae}**	**0.198±0.009^{Be}**	**0.293±0.009^{Ce}**
35	**0.1689±0.0062^{Af}**	**0.286±0.011^{Bf}**	**0.408±0.001^{Cf}**
42	**0.1912±0.004^{Ag}**	**0.348±0.014^{Bg}**	**0.556±0.006^{Cg}**
49	**0.2087±0.002^{Ah}**	**0.418±0.002^{Bh}**	**0.657±0.003^{Ch}**

** The different letters in the table show a significant difference between the values when analyzing by one-way ANOVA with $p<0.05$*

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In order to determine the order of this reaction, we plot the equation PV – t, lnPV – t, 1/PV – t, compare R2 coeficients to determine the order reaction.

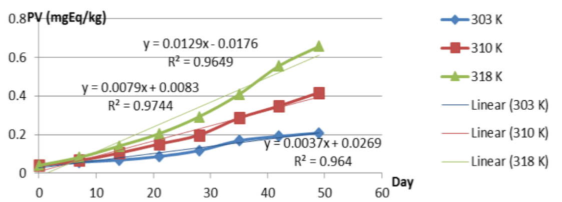

Figure 7. The change of peroxide value over time (PV – t)

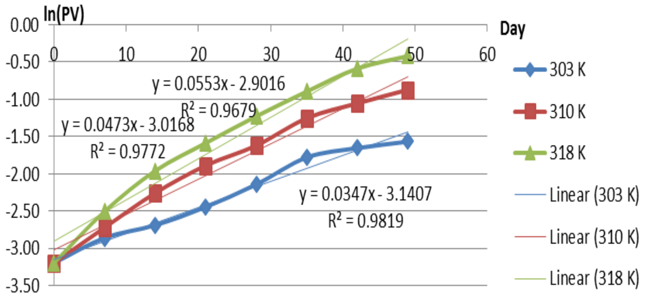

Figure 8. The change of ln(PV) over (lnPV – t)

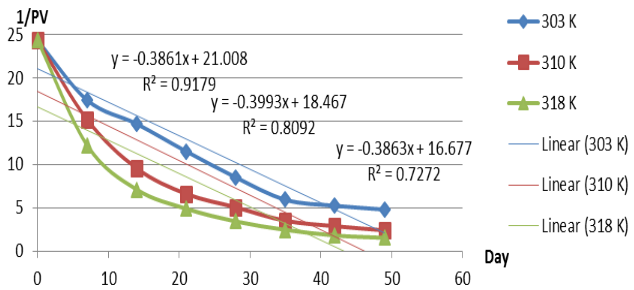

Figure 9. The change of 1/PV over time (1/PV – t)

Due to the R^2 coefficients of the plot in order reaction 1 is the largest, the oxidation reaction of fat is one-order reaction, the reaction rate depends on the reactant concentration.

Table - 5 The reaction rate constant corresponds to the temperatures

T (K)	k	1/T	ln(k)
303	0.0347	0.0033	-3.3610
310	0.0473	0.0032	-3.0512
318	0.0553	0.0031	-2.8950

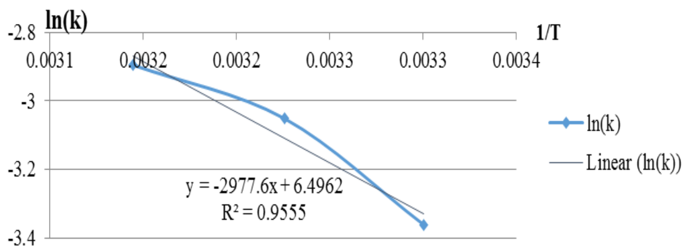

Figure 10. Arrhenius equation plot

The equation lnk = a$\frac{1}{T}$ +b forms lnk = -2977.6$\frac{1}{T}$ + 6.4962, R^2 = 0.9555 (Figure 10). The kinetic parameters for the lipid oxidation rate of mayonnaise include: The energy of activation: Ea = 24.76 KJ / mol; Arrhenius equation constant: k_o = 662.62.

Suppose the actual product storage temperature is 30ºC (303K), lnk = - 2977.6$\frac{1}{303}$ + 6.4962. Inferred k = 0.0358.

According to Russian Customs Union Technical Regulation on fat and oil products, the critical value (A_1) of peroxide value of mayonnaise is 10.0 mEq/kg [18]. The initial peroxide value (A_0) of the sample is0.0411 mEq/kg.

Shelf life of products at 30ºC (303 K) is calculated according to the formula:

$$t = \frac{\ln(\frac{A_1}{A_0})}{k} = \frac{\ln(\frac{10}{0.0411})}{0.0358} \approx 153 \text{ (days)}$$

So, according to the results of peroxide value study, the shelf life of the product if stored at 30ºC is: 153 + 28 = 181 days, completely matches with the manufacturer's published (180 days).

IV. CONCLUSION:

The lipid oxidation reaction is the main damaging factor of the rich fatty product like mayonnaise. The shelf life of mayonnaise stored at 30 ºC predicted according to the Arrhenius equation is 181 days, which matches with the manufacturer's published shelf-life. Hence, we can confirm the accuracy and applicability of mathematical modeling method to quickly predict the shelf life of food products that help producers save time and costs.

Based on the research results, we propose to research using the combination of light and temperature to accelerate the rate of lipid damage to shorten the time for mayonnaise shelf life determination. At the same time, use mathematical modeling for further research on other food products.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest related to this article.

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