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Estimating the Shelf Life of Instant Tempe in Various Packaging and Storage Temperature using the Arrhenius Model

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Abstract

Tempe is a nutritious food that is popular with many people. The problem is that tempe has a short shelf life and must be consumed immediately. Instant tempe is one of the innovative tempe products with a longer shelf life. This research aims to analyze the shelf life of instant tempe in various packages and at different storage temperatures using the Arrhenius model. The method used in this research was laboratory experimental, using 378 samples of instant tempe which were packaged in vacuum packaging, non-vacuum packaging, and cup packaging, and stored at cold temperature (12°C), room temperature (27°C), and hot temperature. (35°C). There are nine treatment combinations regarding the relationship between packaging and temperature, each treatment is carried out 3 times. Measurements for each treatment are carried out until the instant tempe cannot ferment to become tempe ready for consumption. This shows that the fungus Rhizopus oligosporus has died. The research results show that the CIE L value of instant tempe decreases as the storage period for instant tempe increases. The shelf life of estimation results using the Arrhenius model for vacuum packaging and storage temperatures of 12°C, 27°C, and 35°C respectively is 36.98 days, 6.88 days, and 3.00 days. The use of non-vacuum packaging and storage temperatures of 12°C, 27°C, and 35°C were respectively 15.84 days, 5.63 days, and 2.38 days. In comparison, the use of cup packaging and storage temperatures of 12°C, 27 °C, and 35°C respectively are 17.62 days, 6.31 days, and 2.80 days.*

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1. Introduction

Tempe is a native Indonesian food made from soybeans fermented using the fungus Rhizopus oligosporus (Romulo & Surya, 2021; Surono, 2016). Tempe has high antioxidant properties owing to isoflavone aglycones and hydroxylated compounds produced through fermentation (Astawan et al., 2023; Romulo & Surya, 2021). The antioxidant content of tempe is beneficial for maintaining intestinal health, cancer, cognitive function, lung health, heart health, anemia, liver health, bone health, type 2 diabetes mellitus, obesity, skeletal muscle recovery, and malnutrition (Winarno et al., 2021). There are several types of tempes, including mlanding tempe, benguk tempe, gembus tempe, peanut cake tempe, and bongkrek tempe (Romulo & Surya, 2021). However, the tempe currently on the market

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has a relatively short shelf life. At room temperature, tempe can only last two days (Hamzah et al., 2018; Purwanto & Weliana, 2018), even according to (Lastriyanto et al., 2016) the shelf life of tempe at room temperature is less than one day.

Increasing shelf life can be done through packaging-type treatment. The main function of packaging is to protect and protect products from contamination. This includes slowing product damage, extending shelf life, and maintaining the quality and safety of packaged products (Siddiqui et al., 2023). Packaging can also affect the taste, aroma, and texture of tempes. The widely used packaging for tempe is leaf packaging, which can make the shelf life of tempe range between 3 and 4 days (Liuspiani et al., 2020). The use of food-grade plastic packaging can cause the shelf life of tempe to be three days (Santhirasegaram et al., 2016), whereas if it is packaged in vacuum packaging, the shelf life of tempe is four days (Astawan et al., 2016; Razie & Widawati, 2018). The principle of vacuum packaging is to remove all the air from the packaging, then close it tightly so that conditions without oxygen are created in the packaging (Berk, 2018; Lawrence & Kropf, 2024). The absence of oxygen can inhibit the growth of destructive microorganisms and chemical reactions, thereby extending the shelf life of the packaged products (Astawan et al., 2015 ; Kiswanto et al., 2019).

Another factor that influences the shelf life of tempe is the temperature during the fermentation and storage processes. The optimum environmental temperature for the growth of Rhizopus oligosporus is 30°C (Teoh et al., 2024), but other research state that the appropriate temperature for the growth of Rhizopus oligosporus is 35°C (Wahyudi, 2018). Tempes that have been completely fermented can be stored at various temperatures, but storage at 5°C shows the lowest respiration rate and the best quality parameters. The shelf lives of tempe at 5°C, 15°C, and room temperature were 13 days, 5 days, and 2 days, respectively (Purwanto & Weliana, 2018).

One effort to extend the shelf life of tempes is the development of innovative tempe products. Process engineering is carried out to inhibit mold growth during the storage and distribution of tempes by tightly closing tempe packaging. This is because molds grow aerobically and obtain energy from the oxidation of organic substrates (Rahayu & Rahayu, 2021). This tempe product is known as instant tempe. Instant tempe is a tempe product that will be distributed in an unfermented state, so it is still soybeans that have been processed and given yeast, then packaged immediately, so that the soybean fermentation process can be inhibited because the packaging tightly seals the product, preventing it from oxygen. Fermentation of instant tempe is performed when you want to consume it by making holes in the packaging so that the soybeans get oxygen and the tempe mycelium can grow throughout the soybeans. According to (Adit, 2020) this instant tempe product was ranked 1st in the 2020 Indonesia Innovation Award for the Innovative Community Award Category in Devita Noti Wijaya (Central Java) with the title Super Instant Tempe.

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This research aims to analyze the shelf life of instant tempe packaged using various types of packaging and stored at several temperatures. The shelf life was estimated using the Arrhenius model. The Arrhenius model provides a good approximation for describing the temperature dependence of chemical reactions but has also been used for complex biological processes (Crapse et al., 2021). The Arrhenius model can determine the most sensitive parameters during the tempe heating process (Khusnayaini et al., 2018) and the shelf life of tempes stored at cold temperatures and in vacuum packaging (Lastriyanto et al., 2016).

2. Materials and Methods

The ingredients used in this research are soybeans that have been processed into semi-finished tempe or soybeans that have undergone the process of boiling, soaking, removing the soybean shells, washing, and sprinkling with yeast evenly; plastic packaging consisting of LLPDE plastic with a thickness of 90 microns. , and PP plastic cups. The equipment used in this research was a vacuum sealer (YQ-688, Upupin, China), digital scales (I-2000, Kova, China), and a spectrophotometer (ColorFlex EZ, HunterLab, United States). The tool used to predict shelf life was carried out using Microsoft Excel 365 software (2205) and a laptop (EliteBook x360 830 G7, HP, China).

The method used in this research was laboratory experimental, using 378 samples of instant tempe which were packaged in vacuum packaging, non-vacuum packaging, and cup packaging, and stored at cold temperature (12°C), room temperature (27°C), and hot temperature. (35°C). There were nine treatment combinations regarding the relationship between packaging and temperature, and each treatment was carried out three times. Measurements for each treatment were carried out until the instant tempe could not be fermented to become tempe-ready for consumption. This shows that the Rhizopus Oligosporus fungus has died.

Changes in the physical properties of instant tempe during storage were measured by taking pictures of the instant tempe using a camera and mini studio box. Next, instant tempe L*, a*, and b* color testing was performed using a spectrophotometer. The data obtained were processed using Microsoft Excel and presented in a graphical form.

Estimation of shelf life for instant tempe was carried out using the Arrhenius model applied to the Accelerated ShelfLife Test (ASLT) method. The Arrhenius model monitors the effect of the storage temperature on the speed of the degradation reaction of a parameter. The shelf life was estimated using the Arrhenius model by measuring the rate of decline in product quality parameters during storage. The shelf life estimation using the Arrhenius model was performed using the following equation:

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Zero order equation:

$$t = \frac{(C_t - C_o)}{k} \tag{1}$$

First-order equation:

$$t = \frac{\ln\left(\frac{C_t}{C_o}\right)}{k} \tag{2}$$

Where:
t = shelf-life;
Co = quality attribute value at the beginning (day 0);
Ct = quality attribute value at the end (day t); and
k = quality degradation constant.

Validation was carried out for decision-making on the model obtained by estimating the shelf life using the Arrhenius model. The R2 value in each graph was used as the model validation criterion. If the R2 obtained is above 70% (R2≥0.70), then the model is complete, whereas if R2 is below 70 (R2 <0.7) then the relationship between other data needs to be selected (Crapse et al., 2021).

3 Results and Discussion

3.1 Changes in physical properties of instant tempe during storage

During the storage period, the tempe underwent physical changes. The main physical change that can be observed is the color change (Utama et al., 2017). Based on the results of testing the color change of instant tempe using benchtop spectrophotometers, the CIE L* value of instant tempe decreases during the storage period. The decrease in the brightness level of instant tempe during storage was caused by an increase in the number of Rhizopus spp. entering the dead phase, an increase in the amount of linoleic and linolenic unsaturated fatty acids that are susceptible to oxidation, and the formation of red vitamin B12 (Muzdalifah et al., 2017). Changes in the CIE L* characteristics of instant tempe during the storage period are shown in Figure 1.

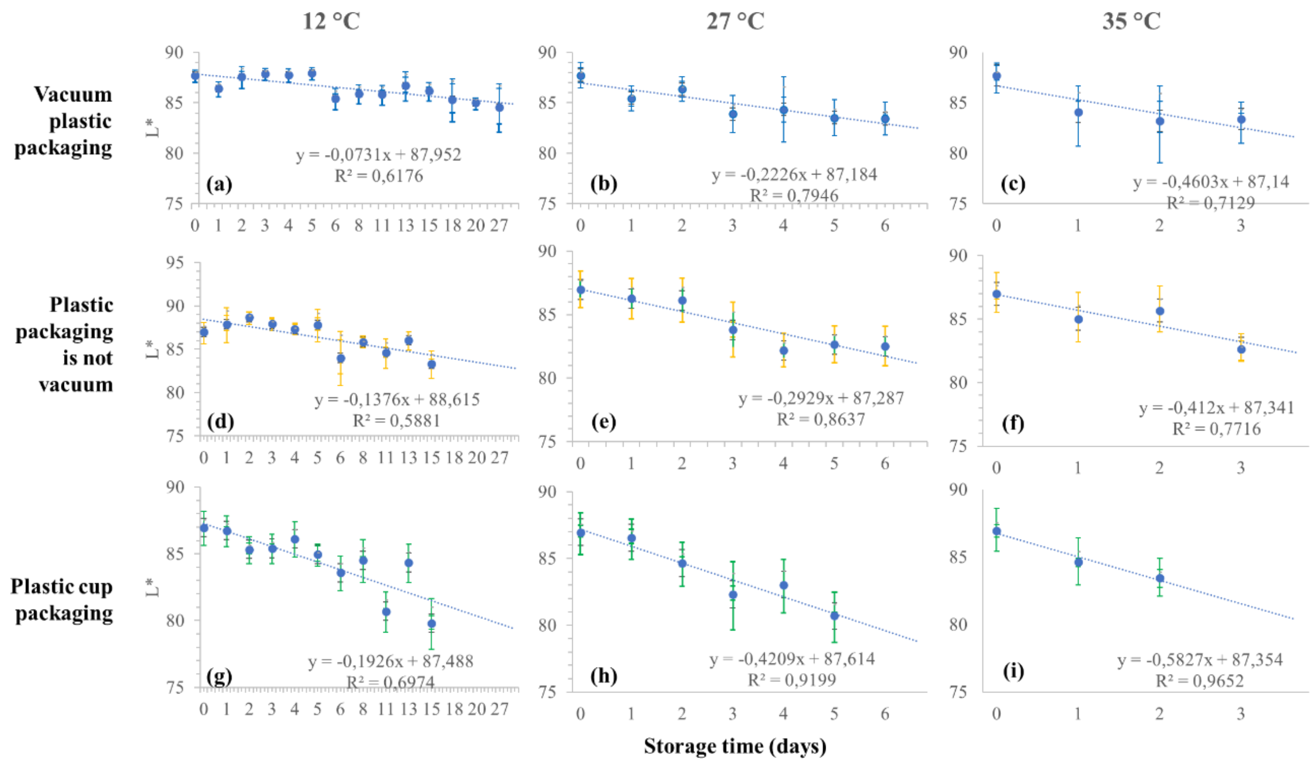

Figure 1. Changes in CIE L* of instant tempe during the storage period at (a) vacuum packaging with a storage temperature of 12 °C, (b) vacuum packaging with a storage temperature of 27 °C, (c) vacuum packaging with a storage temperature of 35 °C, (d) non-vacuum storage temperature of 12 °C, (e) non-vacuum packaging storage temperature 27 °C, (f) non-vacuum packaging storage temperature 35 °C, (g) cup packaging storage temperature 12 °C, (h) cup packaging storage temperature 27 °C, and (i) cup packaging, storage temperature 35 °C.

Figure 1 shows that packaging and storage temperatures reduce the changes in CIE L* values. The rate of decline in the value can be seen from the magnitude of the slope value of the relationship between the CIE L* value and storage time. A negative slope indicated a decline. At a storage temperature of 12 °C, the use of vacuum packaging provides a slope value of -0.1144, which is smaller than that of non-vacuum plastic packaging (-0.2732) and cup packaging (-0.3916). Higher temperature storage provides a greater slope. This shows that the greater the storage temperature, the faster the CIE L* value will decrease, and the use of cup packaging provides the fastest accelerated decrease compared with non-vacuum plastic packaging and vacuum packaging. According to Utama et al. (2017), the type of packaging influences changes in the CIE L* value, and plastic packaging provides the most stable color changes during tempe fermentation compared with leaf packaging.

Another color parameter that indicates a mixed chromatic color of green red is CIE a*. A positive CIE a* value indicates a red color with a range of 0 to +80, whereas a negative value indicates a green

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color with a range of 0 to -80 (Sinaga, 2019). Changes in the CIE a* value of instant tempe during the storage period are shown in Figure 2.

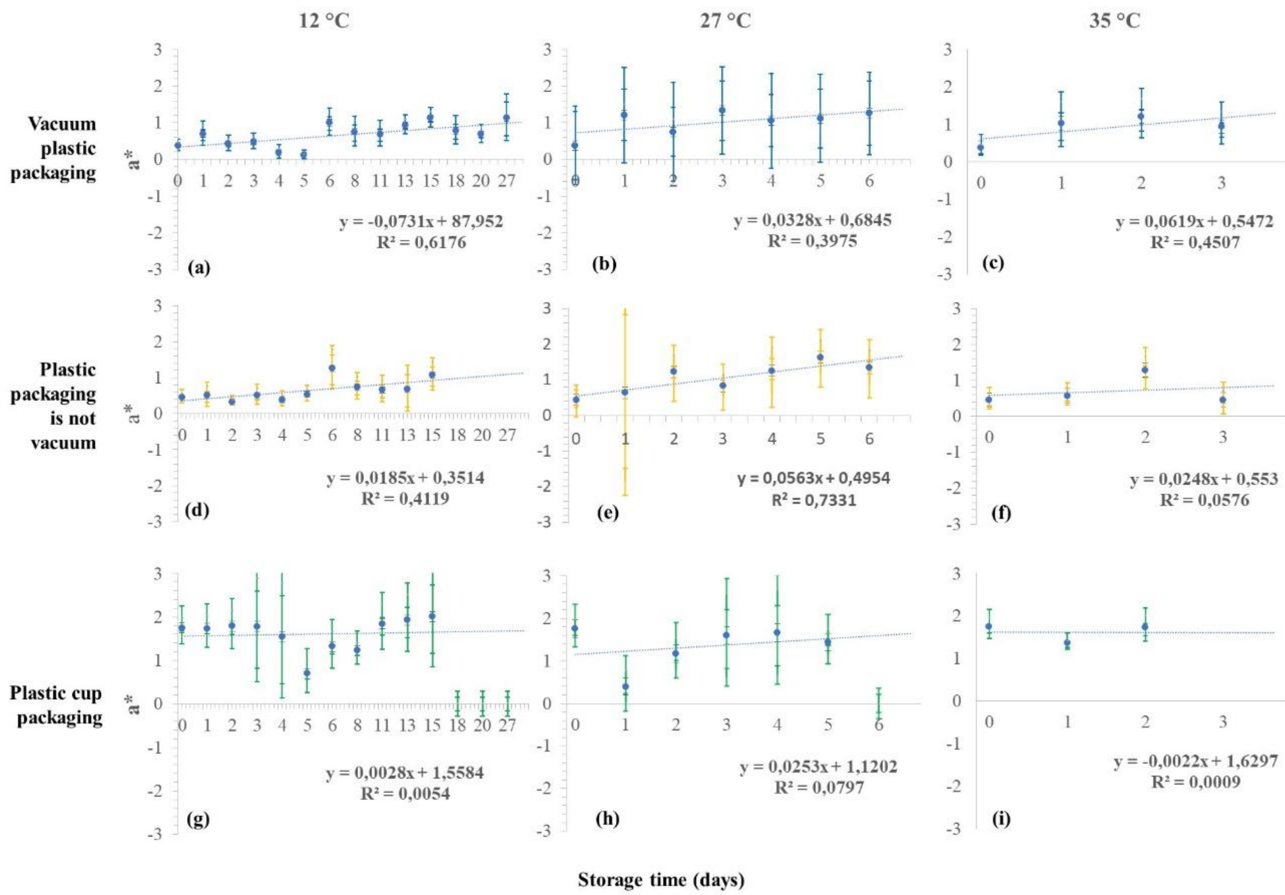

Figure 2. Changes in CIE a* of instant tempe during the storage period at (a) vacuum packaging with a storage temperature of 12 °C, (b) vacuum packaging with a storage temperature of 27 °C, (c) vacuum packaging with a storage temperature of 35 °C, (d) non-vacuum storage temperature of 12 °C, (e) non-vacuum packaging storage temperature 27 °C, (f) non-vacuum packaging storage temperature 35 °C, (g) cup packaging storage temperature 12 °C, (h) cup packaging storage temperature 27 °C, and (i) cup packaging, storage temperature 35 °C.

Figure 2 shows that the test results for the CIE a* value for instant tempe were small, ranging from 0 to 2. This could be because the red or green color indicated by the* value is not the dominant color in the tempe. The entire surface of the tempe is covered by white tempe mold mycelium, which grows during fermentation; thus, the dominant color appearing on the surface of tempe is white (Winarno et al., 2021).

In addition to the CIE L* and CIE a* values, the color that describes the chromaticity of the blue-yellow mixture is CIE b*. A positive CIE b* indicates a yellow color with a range of 0 to +80, whereas a

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negative value indicates a blue color with a range of 0 to -80 (Sinaga, 2019). Changes in instant tempe CIE b* values during the storage period are shown in Figure 3.

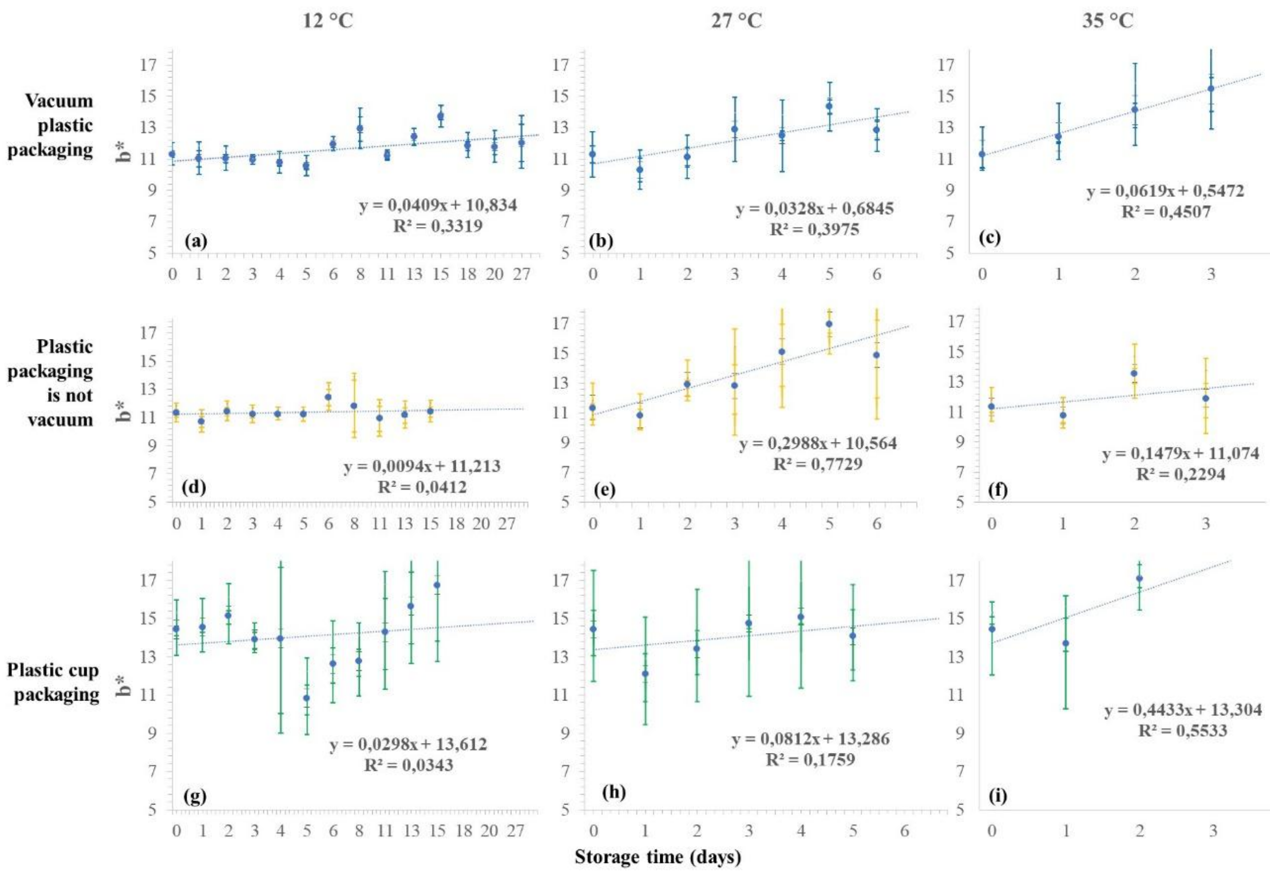

Figure 3. Changes in CIE b* of instant tempe during the storage period at (a) vacuum packaging with a storage temperature of 12 °C, (b) vacuum packaging with a storage temperature of 27 °C, (c) vacuum packaging with a storage temperature of 35 °C, (d) non-vacuum storage temperature of 12 °C, (e) non-vacuum packaging storage temperature 27 °C, (f) non-vacuum packaging storage temperature 35 °C, (g) cup packaging storage temperature 12 °C, (h) cup packaging storage temperature 27 °C, and (i) cup packaging, storage temperature 35 °C.

Based on Figure 3, the CIE b* value increases with the length of storage time, which shows that packaging and storage temperature increase CIE b* (Utama et al., 2017). The CIE b* value obtained shows a value that is not large, and the value has a positive notation (+), which ranges from 11 to 17, where the notation is colored yellow. The yellow color of tempe can be influenced by the raw yellow soybean material (Suhartanti et al., 2019). The b* value, which is not large, could be because the mycelium that grows on the surface of tempe during fermentation is white; therefore, the dominant color on the surface of tempe is white (Muzdalifah et al., 2017; Winarno et al., 2021).

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3.2 Estimation of shelf life of instant tempe

Shelf life estimation was carried out using the order 0 and order 1 equations. This study aimed to obtain a shelf-life estimation model with the best level of validation. The critical parameter determined to estimate the shelf life of the instant tempe in this study is the CIE L* parameter. The CIE L* value was chosen because based on the measurement results, this parameter changes during the storage period and is influenced by the packaging method and storage temperature. In addition, in the tempe fermentation process, the mycelium grows on the surface of the soybean seeds and connects one soybean seed to another so that it becomes a white, compact solid (Razie & Widawati, 2018; Winarno et al., 2021). Mycelium growth can also indicate whether the yeast is still active. If still active, the mycelium can grow evenly throughout the tempe (Abdurrasyid et al., 2023; Muzdalifah et al., 2017), and changes in mycelium growth can be measured using the CIE L* value (Khusnayaini et al., 2018; Utama et al., 2017). The linear regression equations for each storage and packaging temperature are presented in Table 1.

Table 1. Linear regression equation for changes in CIE L* values for instant tempe.

Treatment		Linear regression equation	Slope (k)	Intersep	R^2
Packaging	Storage Temperature (°C)				
Vacuum	12	y = -0,1144x + 87,540	0,1144	87,54	63,50%
Vacuum	27	y = -0,6679x + 86,692	0,6679	86,692	79,47%
Vacuum	35	y = -1,3807x + 86,679	1,3807	86,679	71,29%
Non-vacuum	12	y = -0,2732x + 88,102	0,2732	88,102	58,44%
Non- vacuum	27	y = -0,8787x + 86,994	0,8787	86,994	86,37%
Non- vacuum	35	y = -1,2360x + 86,929	1,236	86,929	77,16%
Cup	12	y = -0,3916x + 86,828	0,3916	86,828	72,72%
Cup	27	y = -1,2629x + 87,193	1,2629	87,193	91,99%
Cup	35	y = -1,7483x + 86,772	1,7483	86,772	96,52%

Table 1 shows that the storage temperature influences the slope (k) value; the higher the storage temperature, the greater the slope (k) value. The vacuum packaging treatment and storage temperature of 12°C had the lowest rate of change in the characteristics (quality loss) because the slope (k) value obtained was the smallest. Based on Table 1, the intercept value for each treatment is quite high because the tempe mold mycelium that grows during the fermentation process is white (Laksono et al., 2019; Winarno et al., 2021), so the CIE L* measurement results show high values in each treatment.

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The relationship between the reaction rate constant and storage temperature can be plotted in the form of ln k versus 1/T (Crapse et al., 2021). Before plotting, the temperature value (T) must first be converted into Kelvin. The ln k and 1/T values were then plotted using an Arrhenius plot. The value of 1/T is plotted on the x-axis, and ln k is plotted on the y-axis. The Arrhenius plots of the CIE L* parameters for each package are shown in Figure 4.

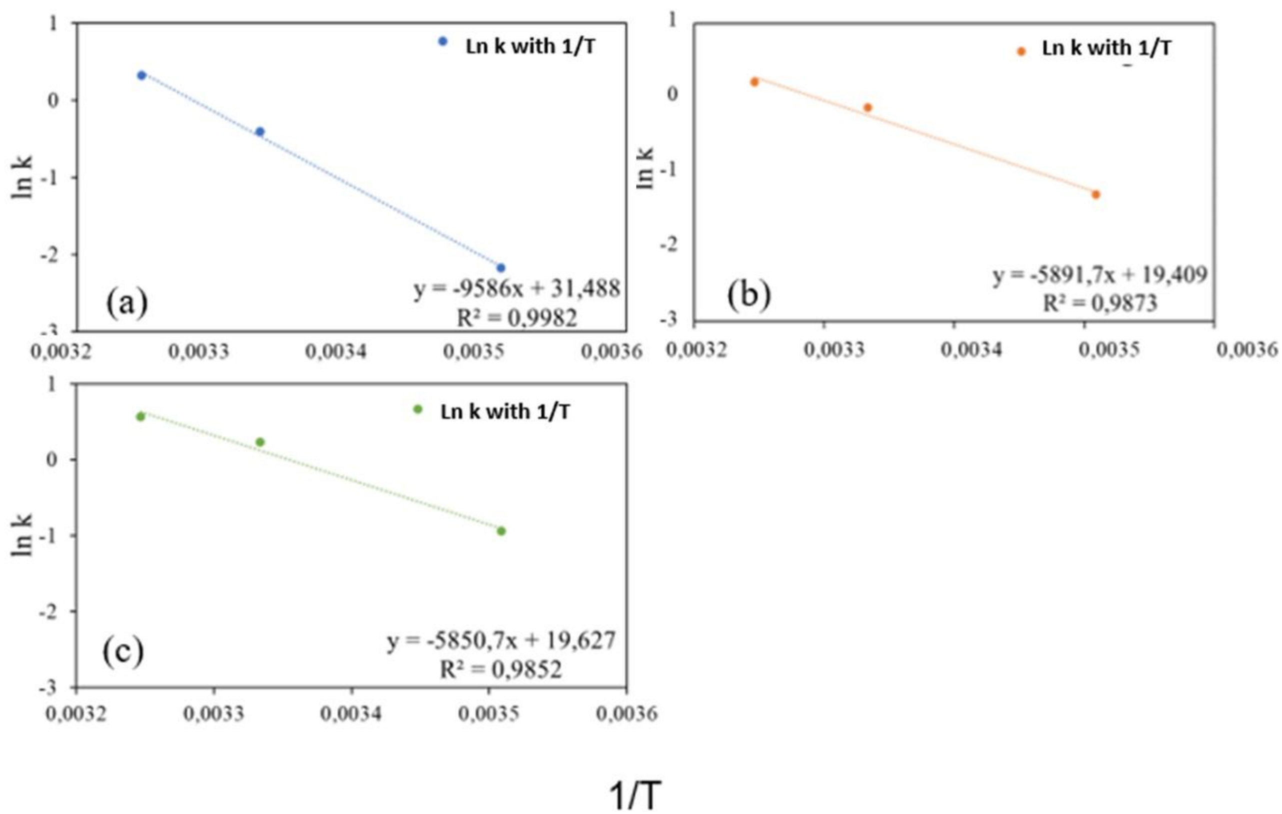

Figure 4. Arrhenius plots of changes in CIE L* in (a) vacuum packaging, (b) non-vacuum packaging, and (c) cup packaging.

Based on the Arrhenius plot of the CIE L* parameters in Figure 4, a linear regression equation for each instant tempe package was obtained. From this equation, we obtain the slope value from the b value and the intercept from the a value. The slope of this equation is the activation energy compared to the gas constant (1.986 cal/mol. K) (Crapse et al., 2021). The intercept value is the ln(k0) value and the exponent of the intercept value is the k0 value.

The constant value of the rate of decline in the quality of the instant tempe (k) can be obtained by calculating the slope, intercept, and 1/T using Equation 1 for each instant tempe packaging. The quality degradation rate constant (k) of instant tempe at each storage and packaging temperature is presented in Table 2.

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Table 2. Constant value of quality degradation rate (k) for instant tempe.

Treatment		k
Packaging	Storage Temperature (℃)	
Vacuum	12	0,117
Vacuum	27	0,628
Vacuum	35	1,440
Non- vacuum	12	0,283
Non- vacuum	27	0,795
Non- vacuum	35	1,323
Cup	12	0,406
Cup	27	1,133
Cup	35	1,880

The k value in Table 2 is the constant rate of decline of the instant tempe quality obtained from the Arrhenius model. This value shows how quality decreases (CIE L*) during storage. The greater the k value obtained, the greater the quality reduction, the greater the quality reduction, the shorter the shelf life (Nursiwi et al., 2021). The lowest k value was obtained in the vacuum packaging treatment stored at 12°C (0.117), which means there was a decrease in the L* value of 0.117 per day.

After determining the constant rate of quality degradation (k) in each treatment, the shelf life of the instant tempe can be estimated. The estimation of shelf life using the Arrhenius model can be obtained using Equation 2. The shelf life based on the estimation results obtained using the Arrhenius model for each treatment is presented in Table 3.

Table 3. Shelf life of instant tempe based on the Arrhenius model.

Treatment		shelf life (days)
Packaging	Storage Temperature (℃)	
Vacuum	12	36,98
Vacuum	27	6,88
Vacuum	35	3,00
Non- vacuum	12	15,84
Non- vacuum	27	5,64
Non- vacuum	35	2,38
Cup	12	17,62
Cup	27	6,31
Cup	35	2,80

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Prediction results using the Arrhenius model show that a storage temperature of 12 °C can extend the shelf life longer than temperatures of 27 °C and 35 °C. The results show that the combination of cold temperature storage treatment (12 °C) and vacuum packaging can maintain an instant tempe with the longest shelf life compared with other treatments. This indicates that the lower the storage temperature, the longer the shelf life of the tempe (Astawan et al., 2023; Purwanto & Weliana, 2018).

3.3 Validation of The Arrhenius Model

Model validation was performed to determine the extent to which the model was represented by the system. The validation of the model for estimating the shelf life of instant tempe for each parameter is presented in Figure 5.

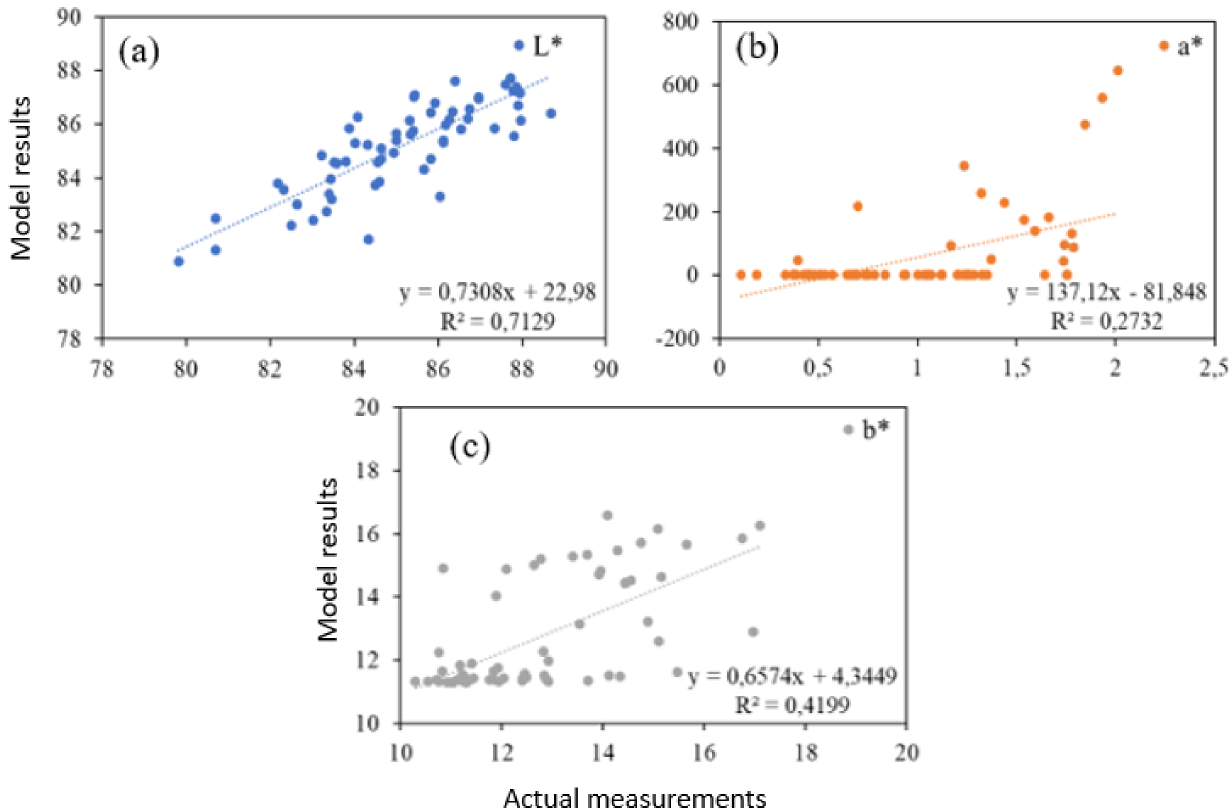

Figure 5. Validation of the Arrhenius model for estimating the shelf life of instant tempe based on parameters: (a) CIE L*, (b) CIE a*, (c) CIE b*

Based on Figure 5, the parameter with the highest model validation results is the CIE L* parameter, with an R2 of 71.29%. The closer the R2 value is to 1 (one), the better the results for the regression model, meaning that the greater the R2 value, the stronger the model will explain the relationship between the independent and dependent variables (Chicco et al., 2021).

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Storage temperature is related to shelf life, and can influence the speed of damage or decrease in quality. Storage at low temperatures can maintain the quality of food ingredients by inhibiting microbial activity and extending shelf life (Astawan et al., 2023; Purwanto & Weliana, 2018). The higher the storage temperature, the greater the reaction speed. A high reaction speed causes a large constant rate of quality degradation (k), such that the product spoils quickly and its shelf life becomes shorter (Lastriyanto et al., 2016). The relationship between storage temperature and shelf life is in line with the research results obtained, where instant tempe stored at a temperature of 12 °C had the lowest rate of quality decline (k value) compared to storage at temperatures of 27 °C and 35 °C. This relationship is also supported by research results (Astawan et al., 2023; Purwanto & Weliana, 2018), which state that the lower the storage temperature, the longer the shelf life.

In addition to storage temperature, packaging also plays a role in the shelf life of instant tempes. The results show that vacuum packaging has a longer shelf life than non-vacuum packaging (non-vacuum and cup). This is because in vacuum packaging all the air is removed from the packaging, so conditions without oxygen are created in the packaging and the growth of damaging microorganisms can be inhibited (Rahayu & Rahayu, 2021) The rate of quality decline in instant tempe with vacuum packaging is smaller than with non-vacuum packaging, so the shelf life can be longer. The relationship between packaging and shelf life is also supported by research results (Astawan et al., 2023; Purwanto & Weliana, 2018; Razie & Widawati, 2018), which state that the combination of cold storage and the use of vacuum packaging can extend the shelf life of tempe compared with other types of packaging.

4. Conclusion

During the storage period, there was a change in the CIE L* value of the instant tempe, where the value decreased as the storage period for instant tempe increased. The decrease in the CIE L* value can be influenced by the growth of mycelia in tempe during fermentation, which decreases with increasing storage time. The shelf lives of the estimation results using the Arrhenius model for vacuum packaging and storage temperatures of 12°C, 27°C, and 35°C are 36.98 days, 6.88 days, and 3.00 days. The use of non-vacuum packaging and storage temperatures of 12°C, 27°C and 35°C were respectively 15.84 days, 5.63 days, and 2.38 days, while the use of cup packaging and storage temperatures of 12°C, 27 °C and 35°C respectively are 17.62 days, 6.31 days and 2.80 days.

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