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Kinetic bio-reaction modelling durian seed fused *Lactobacillus plantarum* suspension by high-order embedded runge-kutta

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Abstract

The advantage and added value of durian seed flour is increased by biochemical modification to substitute the functional properties of wheat flour. The research was carried out by immersing durian seed into *Lactobacillus plantarum* suspension. The purpose was to analyze the effect of durian seed consistency, starter concentration, and several bio-reaction kinetic parameters of durian seed chop with *L. plantarum*. This involves washing and cutting durian seed, immersing the seed in $Na_2S_2O_5$ solution, fermenting durian seed using *L. plantarum* bacteria, drying fermented durian seed, grinding, and sifting 80 mesh of fermented durian seed flour. The modified durian seed sample was analyzed for several functional and microbial parameters, such as the carbonyl group, carboxyl group, degree of substitution, and the number of bacteria by SPC method with various independent variable. The loose variable occurs in 5%, 10%, 15%, 20% and 25% b/v substrate concentration, starter concentration of 2.5%, 5%, 10% v/v and fermentation period initiated from 8 multiples until 40 hrs. The kinetic fermentation model was compiled and optimized solver using a third order runge kutta. Based on the results, modified durian seed flour showed a significant change in functional properties compared to unmodified. The maximum specific growth rate of *L. plantarum* in durian seed slices was 1.902/hr. The maximum bacterial concentrations (Xm), Kd, Yx/s, and Ks obtained in this study were 8.69×10^{20} CFU/mL, 1.631/hr, 5.375 CFU/mL g^{-1}, and 4.49 g/L, respectively. The optimal condition of durian seed flour was at 15% w/v substrate concentration and 5% v/v starter concentration at 24-hr fermentation time with a swelling power value of 8.5 g/g, 7.64% solubility, 162.43% WAC and 28.81% OAC. Though the functional properties obtained were still not close to commercial wheat flour, there was a significant increase compared to native durian seed flour. Hence, the data obtained showed that fermented durian seed flour has potential to be used as wheat flour alternative.

1. Introduction

Wheat flour is one principal food matter in Indonesia. The wheat flour national needs in 2017 reached 11.172 million tons or increased 8.71% on average in a decade (BPS, 2022b). The necessity constantly increased among the Indonesian people over the years. Reducing the import of wheat has encourage more effort in utilizing fruit seed flour in Indonesia resources to substitute wheat flour. Durian seed flour (*Durio zibethinus* Murr.) has potential to be used as a food buffer, likewise it contains 82.04% carbohydrates (Malini *et al*., 2016). In 2021, durian fruit production in Indonesia was 1,353,037 tons and tends to increase year by year (BPS, 2022a). Amin *et al*. (2007) showed durian is usually eaten directly or processed into lunkhead, candy, compote and other products, while the seeds (20-25%) and durian bark are mostly discarded.

Mirhosseini *et al.* (2015) showed unmodified durian seed flour can be used as corn starch alternative in pasta production. It composed of polysaccharide that can improve pasta quality to be more firm and solid dough but not sticky. It is dark color and with strong aroma, making it unbearable by most people. Malini *et al.* (2016) succeeded in replacing 50% tapioca flour composition with durian seed flour in meatballs production. In fact, the durian seed flour adjunct was reported to increase the protein content in meatball. It

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was also used in the cookies and pastries manufacturing (Azima *et al*., 2017). However, glycoprotein compounds in durian seed flour alleviate the chemical and functional properties of durian seed flour. Durian seed flour has several weaknesses, including low gelatinization temperature, baking expansion, water and oil absorption, limited solubility, and sensory properties which were not acceptable to the public. These constraints made durian seed flour still unusable in the food industry. Therefore, durian seed flour quality improvement is important by modifying its functional properties either physically, chemically or biologically.

Rectifying the durian seed flour physicochemical trait have been carried out by several researchers. Soaking durian seed flour in sodium metabisulfite solution has been shown to improve the physicochemical properties of the flour (Kumoro and Hidayat, 2018). Zuhri *et al.* (2015) reported that durian seed flour fermentation using tape yeast can produce modified flour with moisture content, ash content, and baking expansion which almost equivalent to commercial wheat flour. Jayus *et al.* (2016) showed that using *L. plantarum* for jackfruit seed flour fermentation can reduced oligosaccharide (stachyose and verbacose) that caused human tummy puffed up. Fermentation action by *L. plantarum* succeeded to change starch structure, amylopectin and amylosa structure, and some functional properties (Hashemi *et al*., 2017; Rahma *et al*., 2017). Consequently, fermentation technology was applied in modifying durian seed flour as food intermediate. The fermentation technology development is based on the fact that the Indonesian people are used to processing foodstuffs with fermentation techniques, such as in the manufacture of tempeh or oncom, soy sauce, tempoyak and tape.

This study set out to determine the impact of solid consistency (5–25% w/v), starter concentration (2.5–15% v/v), and fermentation time (0–40 hrs) on the degree of substitution, carbonyl and carboxyl group content while fermenting durian seed chips with *L. plantarum*.

2. Materials and methods

2.1 Plant materials, microorganism and chemical

This research used durian (*Durio zibethinus* Murr.) seed as the main material to obtain the flour product. The durian seed was collected from Indonesian farmers in Balikpapan. The fruit pulp was cleaned and washed with flowing water. The seeds were dried in drying oven at ambient temperature (30ºC), collected in plastic bags and kept in dry and cool condition at 5ºC for 24 hrs (Al-Baarri *et al*., 2020). Sodium metabisulfite as a preliminary addition material to prevent browning reaction (Kumoro and Hidayat, 2018) was obtained from authorized chemicals distributors in Balikpapan. *Lactobacillus plantarum* CCRC 12251 as the fermenter agent was bought from Food and Nutrition Universitas Gadjah Mada, Yogyakarta, Indonesia. The bacteria were maintained in Mann Rogassa Sharpe (MRS) agar slant at 4ºC. All of the chemical reagent were of analytical grade purchased from chemical distributors in Balikpapan.

2.2 Inoculation preparation

Erlenmeyer flask contain 100 ml of modified MRS liquid (20 g glucose, 10 g peptone, 10 g beef extract, 5 g yeast extract, 5 Na_2HPO_4, sodium acetate, 2 g triammonium citrate, 0.2 g $MgSO_4$, 0.2 g $MnSO_4$, 4 g $CaCO_3$ 4 L^{-1}, 0.1 mL Tween 80 and pH 6.8) was inoculated using *L. plantarum* from a stock culture, and incubated at 35ºC and shaking at 120 rpm for 48 hrs in an orbital incubator shaker. The number of bacteria was calculated with standard plate count (SPC) method as suggested by Widayat *et al*. (2020).

2.3 Durian seed chip fermentation

Durian seeds were chopped to get chips shape with 5 mm thickness. Preliminary addition of 0.6% (w/v) $Na_2S_2O_5$ was to prevent durian seed browning reaction for 2 hrs (Kumoro and Hidayat, 2018). Pour 200 ml of distilled water into 500 mL Erlenmeyer flask, then multiform solid consistency (5-25%) (w/v) were achieved. It was inoculated with varying values of freshly provided inoculums (2.5-15%) (v/v) and covered with aluminium foil. Thermal sterilization was not performed to prevent starch gelatinization. Ensuring adequate association and temperature oversight (35ºC), the flask containing the durian seed fermented was placed on a horizontal shaker water bath. After fermentation periods of 8, 16, 24, 32 and 40 hrs, fermented durian chips were collected. They were rinsed directly with flowing water and then dehydrated on mono layer arrangement drying pans in an electric oven at 40ºC for 3 days. A hammer mill was used to pulverize the dried chips, then to obtain finer flour used a ball mill. Subsequently, the flour was sieved through 180-250 um screens to remove unwanted particles. Polyethylene plastics bags were used to store the durian seed flour, which was then placed in plastic containers at 20ºC for proper material preparation. These containers were utilized for further analysis, following the recommendations of Retnowati *et al.* (2018).

2.4 Raw materials and product elucidation

2.4.1 Chemical properties

The proximate composition of all durian seed flour samples was analyzed by following the official method

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(Latimer, 2016). The carboxyl content was determined as previously described (Fonseca *et al*., 2015). The carbonyl content was determined conforming to the titrimetric hydroxylamine method (Klein *et al*., 2014). Both of them were measured with some modification. The degree of substitution (DS) was measured following Sodhi and Singh (2005) method titrimetrically.

2.4.2 Statistical analysis

All data in triplicates were acquired as mean ± standard deviation (SD). Significant differences between the mean values at level $p<0.05$ were compared using Microsoft Excel 2021 MSO (Version 2207 Build 16.0.15427.20060) 64-bit.

2.4.3 Modelling apparatus

Calculating the number of *L. plantarum* during fermentation process was done by standard plate count (SPC) method with some modification (da Silva Sabo *et al*., 2017). *Lactobacillus plantarum* kinetic parameter use first-order Gombertz model (Wardhani *et al*., 2019). Third-order Runge-Kutta resolve the formulation. Then proceeds by comparing between the Logistic, Richards, and Baranyi Model for feasibility forecasting. Maximum bacterial unit at suspension (Xm) and death specific rate (Kd) was calculated by the Monod equation (Ray, 2004). Mass substrate in the suspension was profiled in Lineweaver-Burk diagram by the method described by Mitchell *et al*. (2004). All kinetic parameter were modelled and profiled using Microsoft Excel 2021 MSO (Version 2207 Build 16.0.15427.20060) 64-bit.

3. Results and discussion

3.1 Effect of substrate concentration depends on carboxyl, carbonyl and degree of substitution

Durian seed have been soaked in 0.6% w/v $Na_2S_2O_5$ solution to prevent the browning reaction. After that, it was fermented using *L. plantarum* by submerged batch process. Optimizing the weight consistency of durian seeds is necessary to modify the operational conditions using the fermentation method with *L. plantarum*. The consistency of durian seeds is closely related to *L. plantarum* as it produces the amylase enzyme. This enzyme has the capability to react with substrate, modifying the starch granules and thereby increasing the functional properties of durian seed flour.

The carboxyl group increased by 62.14% from the durian seed consistency from 5% until 15% (w/v). It occurred due to the activity of *L. plantarum* which was characterized by increased extracellular amylase, proteases and tannase enzymes production (Kannan *et al.,* 2011; Wardhani *et al*., 2022). It can be concluded that the more substrates available, the more bonds and reactions happening between enzymes and substrates (Rogers and Gibson, 2009). When the durian seed consistency exceeded 15% (w/v), there was a decrease in the carboxyl group content by approximately 20.82%. This decline in carboxyl group content can be attributed to the gradual dehydration of *L. plantarum*, leading to a reduction in bacterial activity. The decrease in bacterial activity was caused by an excessive amount of substrate available for bacterial growth. In cases of extremely high substrate concentration, cell dehydration occurs in the concentrated solution, inhibiting the fermentation process (Silviana *et al*., 2021).

As the substrate concentration livens up, the carboxyl content output decreased. A higher solid consistency will lead to an increase in culture medium viscosity, resulting in a reduction in water activity and inhibition of bacterial growth. Wardhani *et al.* (2009) showed that the fermentation system has changed from liquid (submerged fermentation) to semi-solid or solid (solid state fermentation). In agreement with Aryanti *et al.* (2017), bacteria generally thrive at high water activity or moisture content. The result of this study also indicated a correlation between the amount of carbonyl content and a substrate concentration of 15%, as shown in the graphs (Figures 1-9). This provide further evidence that bacterial activity was high during that time. The carbonyl group is formed as a result of *L. plantarum* activity, where it oxidizes the hydroxyl group in the starch chain of the durian seed.

There was an increase in the levels of both the carboxyl groups (53.8%) and carbonyl groups (75.2%) in 25% substrate concentration (w/v). The remaining carbonyl content oxidation in the fermentation system was observed. This phenomenon occurred at a substrate concentration of 25% (w/v), but the measured carbonyl content was only 0.233%. This value was lower than the carbonyl group content in the control variable, which was 0.27%. Despite the escalation in both carbonyl and carboxyl content, it can be concluded that there was a lack of *L. plantarum* activity. The bacteria consumed the excess substrate which caused the fermentation system to turn into semi-solid or solid state fermentation. This is in accordance with the suggestion put forward by Wardhani *et al*. (2009).

The DS value decreased in all variables as a consequence of *L. plantarum* bacteria inhibition. This inhibition occurred due to a slight concentration of the culture medium bacteria, which blocked cell growth (Silviana *et al*., 2021).

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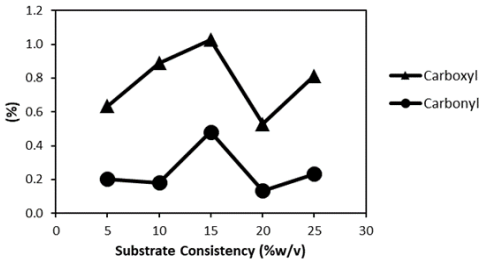

Figure 1. Influence substrate concentration for carbonyl and carboxyl content.

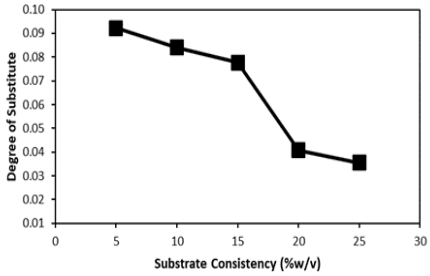

Figure 2. Influence substrate concentration for degree of substitution.

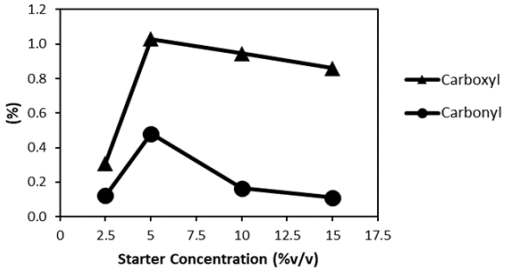

Figure 3. Influence starter concentration for carbonyl and carboxyl content.

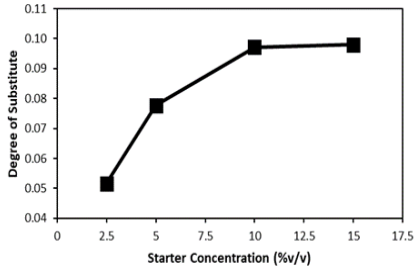

Figure 4. Influence starter concentration for degree of substitution.

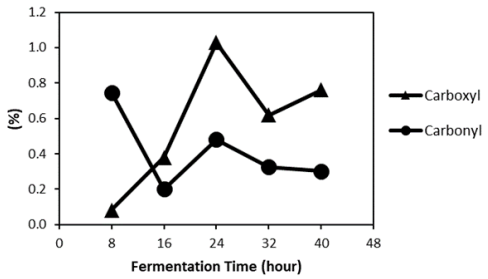

Figure 5. Influence time allocation for carbonyl and carboxyl content.

Disrupted cell growth will cause OH groups inhibition which could be substituted by lactate groups produced by *L. plantarum* (Soumya *et al*., 2019). Therefore, it can be deduced, the more the durian seed consistency, the lower the hydroxyl group that is replaced due to the lack of *L. plantarum* activity in immersioning fermentation system, revealing a decrease in both levels of carboxyl and carbonyl content when the substrate concentration was intensified during durian seed fermentation.

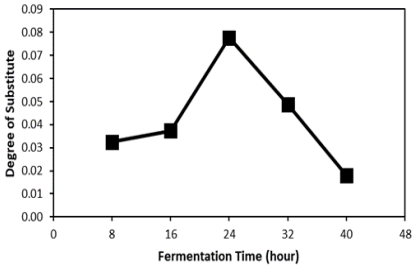

Figure 6. Influence time allocation for degree of substitution.

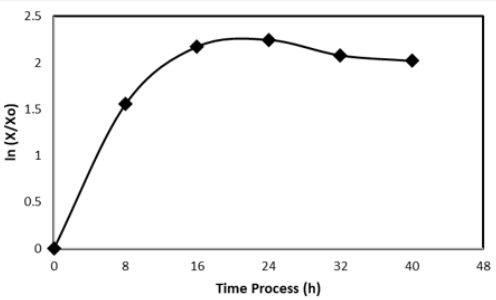

Figure 7. *L. plantarum* growth rate by SPC method.

3.2 Effect of starter concentration depends on carboxyl, carbonyl and degree of substitution

Besides substrate concentration, the *L. plantarum* suspension is also important parameter to obtain high yields and productivity in submerged fermentation system. At low starter concentrations, the substrate is used slowly thereby prolonging the incubation time. Meanwhile, at high starter concentrations, it can cause growth competition between microorganisms with limited substrate number (Nagarjun, 2015). The effect of microorganism growth cannot be separated from the levels of carbonyl and carboxyl groups.

The research data exhibited increasing level of the carboxyl group significantly from 0.309% to 1.028%. This was due to escalating in *L. plantarum* activity. It was also followed by the carbonyl group content from 0.48% to 0.12% at the same concentration. The rising activity due to the activity of the various enzymes such as the amylase, protease and tannase (Kannan *et al*., 2011; Wardhani *et al*., 2022). It can be deduced that when more starter is available, there are more reactions between enzyme and substrate, resulting in the formation of lactic acid.

The lactic acid formation is initiated by amylase enzyme which was produced from the *L. plantarum* metabolic process. Afterwards, the amylase enzyme ruins the starch structure on amylose molecule to become

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semi-crystalline (Wardhani *et al*., 2022) and provide space for lactic acid to substitute hydroxyl groups with lactate groups. After the substitution process, it is followed by amylopectin linearization. Strengthening over the bonds between starch molecule can improve the durian seed flour's functional properties. Lactic acid production is induced by the pyruvic acid reduction by nicotinamide adenosine dinucleotide hydrogen (NADH) from the glucose breakdown (substrate) which is also known as the glycolysis process (Sieuwerts *et al*., 2018). However, both the carboxyl and carbonyl groups decreased significantly after the starter concentration was more than 5%. The alleviation of the carboxyl group by 16.34% and the carbonyl group by 65.83% was caused by an increase in culture medium viscosity. As a result, the water activity decreased, leading to the inhibition of the growth and development of *L. plantarum* bacteria. Therefore, the fermentation system changed from liquid (submerged fermentation) to semi-solid or solid state fermentation (SSF).

The DS value increased at each starter concentration variable, and even the tendency of the starter concentration above 10% remained relatively constant. The increase in the degree of substitution value is attributed to *L. plantarum* being in the lag phase (adaptation) and exponential phase, where the number of living bacteria increased rapidly. Consequently, the OH groups can be substituted by the lactate group from the *L. plantarum* outcome.

The longer the reaction time, the more lactate groups will diffuse and adsorbed through the starch surface (Soumya *et al*., 2019). However, when the starter concentration is above 10%, the degree of substitution number is relatively constant since the lactic acid production did not show significant change (Tripathi *et al*., 2015). This indicated that microbial growth is in stationary phase.

3.3 Effect of time fermentation depends on carboxyl, carbonyl and degree of substitution

In lag phase (adaptation phase), before 16 hrs the carbonyl group content decreased significantly by 73.09%. This is due to the carbonyl group formed during incubation in agar media is oxidized to become carboxyl group until the fermentation time is 16 hrs. After 24-hr, increasing carbonyl content (32.39%) is caused by *L. plantarum* improvement activity. It formed carboxyl group which were oxidized directly to carbonyl group again. After 24 hrs of fermentation, the carbonyl group content decreasing and remains constant. Since the bacteria begin to be in the death phase experience, they cannot be reactivated to implement oxidation reaction. These results are in agreement with Putri *et al*. (2012), in which the hydroxyl group on starch molecule is made from first carbonyl group oxidation and then further oxidized to be carboxyl group as the final product until death phase.

Along with the fermentation process, the levels of carboxyl groups (-COOH) contained in modified durian seed flour was also changed. It keeps increasing for up to 24 hrs. The number of carboxyl groups increasing occurred due to further carbonyl group oxidation by *L. plantarum.* The hydroxyl group in starch were oxidized to a carbonyl group during fermentation. Hereafter, that carbonyl group will be oxidized to carboxyl group which is the final product conversely (Putri *et al*., 2012). Although there was decreasing at 32 hrs fermentation time, it is due to bacteria in death phase position which resulted with decreasing carboxyl and carbonyl groups number. At 40 hrs of fermentation, there was slight increase in carboxyl group content (22.9%) that was the residual oxidation product. On the other hand, the amount of carbonyl groups decreased and the remain tended to remain unchange because *L. plantarum* were no longer able to oxidize the hydroxyl groups on substrate.

The DS value increased during the fermentation time (0-24) hrs and then gradually decreased until 40 hrs fermentation. The DS value escalating occurred when *L. plantarum* experienced in lag phase (adaptation) and exponential phase. The longer the reaction time, the more lactate groups will diffuse and adsorbed through the starch surface. However, during the time when the DS value is lower, *L. plantarum* bacteria have experienced a death phase, and the number of viable bacteria also decreases.

Thus, the amount of OH groups can be substituted by lactate groups from lactic acid delivered by bacteria. Furthermore, the longer fermentation time can trigger the hydrolysis of lactic starch (Yaqin *et al*., 2019). Even though, overall, the lactic acid produced from modified durian seed flour was suitable for use as food. Liu *et al.* (2022) asserted the lactic acid starch with a DS value of around 3 is considered suitable for consumption.

3.4 Lactobacillus plantarum bacteria count in durian seed consistency profile and modelling

Lactobacillus plantarum bacteria growth while in fermentation process was calculated by SPC method. Elsewhere, some lactic acid bacteria for growth phase and its ability to produce lactic acid was performed in batch process (Hashemi *et al*., 2017). Generally, bacteria growth consists of lag phase (adaptation), exponential phase, stationary phase, and death phase. This result can be evidenced by sum of *L. plantarum* bacteria

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improvement.

However, lag and stationary phase weren't observed in details in this research. The longer the fermentation process, thus the more bacteria grow in durian seed suspension. Kedia *et al*. (2008) reported the phenomena in which wheat flour fermented with *L. plantarum* show that at 24 hrs process, the bacteria were in the peak of growth schema. The number of bacteria was 7.5 CFU/ mL. Following this growth, the bacteria growth slow down until death phase. On the other hand, Hashemi *et al.* (2017) reported that the fermentation process of lemon juice in *L. plantarum* within the 0-24 hrs timeframe significantly improved the growth of the bacteria. However, after 36 hrs, the fermentation process entered the stationary phase. In this study, the bacteria were in the lag phase within the first 8 hrs, as they adapted to the nutrition in the media. Therefore, sampling within a shorter time frame (0-1) hr should be done to account for *L. plantarum*'s lag phase more accurately in the durian seed media. Subsequently, the exponential or logarithmic phase lasted until the 24-hr process. Comparing with the exponential phase of *Lactobacillus bulgaricus* and *L. casei* (Taleghani *et al*., 2016), which began at 36 and 48 hrs, respectively. After the logarithmic phase, the stationary phase followed, where the bacteria experienced constant growth between life and death. During this phase, the bacterial cells did not reproduce as the nutrition concentration in the logarithmic phase decreased. The transition from the exponential to the stationary phase was delayed because the cell growth rate in the culture was faint, even before the substrate was significantly consumed. However, this research did not show the stationary phase. After stationary phase, the bacteria cells died due to a lack of nutrients. The accumulation of acid and waste metabolites since the 32-hr fermentation process indicated that the bacteria could not survive as long as *L. casei* and *L. bulgaricus* (Taleghani *et al*., 2016). This phenomenon was not consistent with previous studies (Luz *et al*., 2020; Sun *et al*., 2022), which reported a death phase in lactic acid bacteria after incubation later, around 72 hrs. The kinetic growth divination using the first-order Gombertz equation (Wardhani *et al*., 2019).

$$\frac{dy}{dt} = A\,\lambda\,.\,exp[exp(\mu_m - \lambda\,t)].\exp\,(\mu_m - \lambda\,t)$$

Where A is asimtot, um is specific bacteria growth rate maximum, and lamda is lag phase in hr. Completion of Gombertz equation was using third-order Runge Kutta for the value of three parameter. The following third-order of Runge Kutta composed of:

$$k_1 = hf\,(x_r, y_r)$$

$$k_2 = hf\left(x_r + \frac{1}{2}h, y_r + \frac{1}{2}k_1\right)$$

$$k_3 = hf(x_r + h, y_r - k_1 + 2k_2)$$

$$y_{r+1} = y_r + \left(\frac{k_1 + 4k_2 + k_3}{6}\right)$$

The graphic (Figures 8 and 9) indicates a deviation value of 0.0016, suggesting that the Gompertz model is the most suitable among the others. The kinetic parameter values were determined to be um 1.902/hrs, and the lag phase lasted approximately 0.342 hrs. Subsequently, the Logistic method (Kedia *et al*., 2008) was used for mathematic modelling to compute Xm (CFU/mL). The values of Xm were found to be (8.69×10^{20}) CFU/mL with a deviation of 3.589 aberration value.

$$X = \frac{X_m}{1 + \exp\left[2 + \frac{4\,\mu_m}{X_m}.(\lambda - t)\right]}$$

Thereafter, specific death rate (Kd) in hr was using Monod model (Rogers and Gibson, 2009) with boundary condition t = 0 then X = X_0, at t = t subsequently X = X. The linearization of differential equation can be transformed to:

$$\frac{dX}{dt} = (\mu - kd)X$$

$$\ln X = \ln X_0 + (\mu - Kd)t$$

Substitution of u value that referral by Spier *et al.* (2009) measured the Kd value 1.631/hr.

$$\ln X = \ln X_0 + \left[\mu_m\left(1 - \frac{X}{X_m}\right) - Kd\right].t$$

The yield coefficient (Yx/s) for biomass and substrate concentration (Ks) were calculated using the Monod equation modified by Mitchell *et al.* (2004). The Lineweaver-Burk graphic was utilized to decipher the formula with a slope of 1/u and 1/[S]. From the slope, Ks was found to be 4.49 g/L, and um was determined to be 1.902/hr based on the Gompertz model.

The optimization of kinetic parameters provided the specific maximum growth rate of *L. plantarum*. In a previous study, Pramono *et al.* (2003) calculated the *L. plantarum* growth rate in glucose media at 0.26/hr, while Horn *et al.* (2005) reported specific growth rates of 0.6/ hr in fish viscera hydrolysis media and 0.4/hr in seaweed media. The specific growth rate obtained in this research is higher than the previous studies, which can be attributed to the durian seed suspension having more complex substrate molecules compared to glucose. Glucose is a monosaccharide with relatively modest atom-C and bon complexity. Furthermore, other important parameters include the maximum bacterial unit in suspension (Xm) at (8.69×10^{20}) CFU/mL, death specific rate (Kd) at 1.631/hr, yield coefficient (Yx/s) for biomass production at 5,375 CFU/mL g^{-1}, and substrate

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concentration (Ks) while in specific growth rate equality at 4.49 g/L.

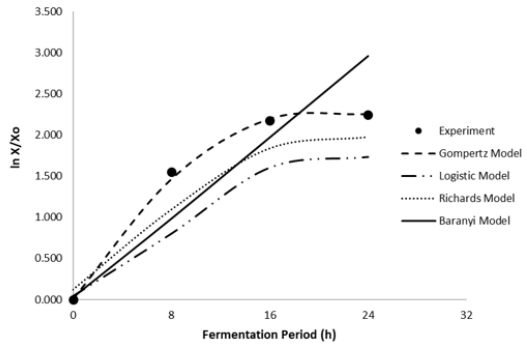

Figure 8. Growth model fermentation by *L. plantarum* in vitro experiment.

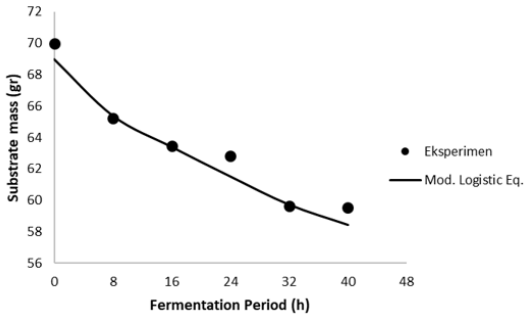

Figure 9. Substrate degradation model by *L. plantarum* in vitro experiment.

3.5 The functional properties of durian seed flour at optimum condition

The functional properties of durian seed flour are intensified as a result of the oxidation process with sodium metabisulfite solution, leading to wider starch granules. Subsequently, the fermentation process maximizes the improvement of functional properties, including swelling power (SP), water solubility (WS), water absorption capacity (WAC), and oil absorption capacity (OAC), resulting in values of 8.5 g/g, 7.64%, 162.43% and 28.81%, respectively. In general, the functional properties of fermented durian seed flour improve and greater than those of the native samples, but they are still lower than those of commercial wheat flour, which has SP, WS, WAC and OAC values of 9.3 g/g. 8.5%, 253% and 195%, respectively. The OAC is still much lower than the standard for wheat flour (195%) due to the low fat content in durian seed. It can be concluded that fermented durian seed flour is not suitable for making dough which is fried by oil to form martabak and odading. The amylose content in the flour binds water molecules directly rather than fat compounds, resulting in minimal changes to fat content after the fermentation process. The absence of fat in durian seed flour increases its ability to blind, expand, and dissolve in water due to the hydrophilic nature of the amino acids chains in the protein (Segura-Campos *et al*., 2015; Shiraga *et al*., 2016). From a molecular perspective, the carboxyl group replacing the hydroxyl group increase the WAC percentage due to the fermentation process. The lactate group, which is a carboxyl group with a hydroxyl group attachment, becomes soluble in alcohol and hygroscopic matters. These processes accelerate the movement of durian seed molecules, allowing more water to enter inside the pores (Coral *et al*., 2009).

The swelling power values of jackfruit seed starch, cempedak seed and Thai durian seed are 8.61 g/g, 4.56 g/g and 11.95 g/g, respectively. Meanwhile, the solubility values are 17.08% (Mukprasirt and Sajjaanantakul, 2004), 9.56% and 21.64% (Baraheng and Karrila, 2019), respectively. The increase in some functional properties is caused by a reduction in amylose content during the fermentation process by *L. plantarum*. Amylose is stripped towards the outside of the granules to form a gel -shaped matrix that envelops the starch granules containing mostly amylopectin. Additionally, the amylase enzyme produced by *L. plantarum* can break the amylose and amylopectin bonds (amylopululanase).

Lactic acid biologically remove amylose from the surface of flour crystals and change its physical properties to make it more amorphous (Senanayake *et al.*, 2013; Wardhani *et al*., 2022). Indeed, the acidic culture conditions and high enzyme activity induced amylose to undergo further hydrolysis so amylose level fell. In line with the research on corn flour fermentation by other researchers (Anasiru *et al*., 2019; Ma *et al*., 2022; Wei *et al*., 2022). The amorphous starch structure in durian seed flour leads to the fermentation reaction converting complex carbohydrates (polysaccharide) into simple carbohydrates (monosaccharides). Consequently, a higher substate concentration escalates the amylase enzyme activity and increase the flour's porosity. This transformation result in improved functional properties of durian seed flour, such as increased swelling power (SP), water solubility (WS) and oil absorption capacity (OAC) (Zuhri *et al*., 2015; Wardhani *et al*., 2022). However, these functional properties are still below the standard of commercial wheat flour (Chung *et al*., 2010).

4. Conclusion

The 15% substrate concentration in fermented media is optimum result to modify durian seed by *L. plantarum*, with carboxyl content of 0.48% and carbonyl content of 1.028%. The degree of substitution, swelling power, solubility, water absorption capacity (WAC), and oil absorption capacity (OAC) were determined to be 0.078, 8.5 g/g, 7.64%, 162.43% and 28.81%, respectively. The optimum fermentation time was found to be 24 hrs, significantly influencing the functional properties of the modified durian seed flour. Ultimately, durian seed flour

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fermented by *L. plantarum* exhibited higher swelling power, solubility, WAC, and OAC compared to the unmodified version. However, the WAC dan OAC traits of the modified durian seed flour were still significantly lower than those of commercial wheat flour.

Conflict of interest

The authors declare no conflict of interest.

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