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Synthesis of Lactic Acid from Molasses by *Lactobacillus acidophilus* Using a Batch Fermentation Process

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Abstract

Lactic acid is a chemical with widespread applications, mainly in pharmaceutical, cosmetic, chemical as well as food industries. One of the major uses of lactic acid is the precursor of polylactic acid (PLA). PLA is biodegradable and biocompatible material that can be an alternative to conventional plastic derived from fossil fuels. Efforts continue to be made to reduce the high cost of producing PLA in order to compete with conventional petrochemical-based plastics. One of which is the use of molasses as a raw material because it is cheap and contains high quantity of glucose. This study aims to obtain the value of the cell concentrations with various addition of starter volume, that produces high concentrations of lactic acid, and to obtain the growth kinetics of *Lactobacillus acidophilus* during the fermentation process. This study was conducted in several stages, i.e., the design of bioreactors, inoculation of *Lactobacillus acidophilus*, and fermentation of molasses. In a batch system, molasses substrate was directly inserted as much as 500 ml into the bioreactor and various amount of starter of *Lactobacillus acidophilus* (1; 3; 5 dan 10% v/v) were added to the fermentation medium. The fermentation of molasses observed for 72 hours and the product was analysed every 8 hours. The highest concentration of lactic acid produced in batch fermented molasses, 23.1 mg/L, resulted from the addition of 5% (v/v) starter bacteria, that last for 72 hours fermentation time. The value of the carrying-capacity coefficient (k) and the maximum net specific growth rate (μ_{net}) were 0.2379 h^{-1} and 0.0160 h^{-1} respectively.

Keywords: molasse, lactic acid, *Lactobacillus acidophilus*, fermentation

1. Introduction

Lactic acid (2-hydroxypropionic, $CH_3CHOHCOOH$) is an important chemical and is widely used in the cosmetic, pharmaceutical, chemical, textile and food industries. One of the most promising applications of lactic acid is the use of lactic acid as a raw material for the manufacture of polylactic acid (PLA). PLA is a versatile, biodegradable, aliphatic polyester polymer derived from 100% renewable resources (Abdel-Rahman et al., 2011; Komesu et al., 2017). PLA is environmentally friendly, and can be decomposed into water and carbon dioxide by microbes (Li et al., 2020). PLA can be obtained from lactic acid through various polymerization processes such as polycondensation, ring opening polymeri-zation, azeotropic dehydration, and enzyme polymerization (Savioli Lopes et al., 2012).

Lactic acid, as the monomer of PLA, can be produced through fermentation and chemical processes. Fermentation is the most widely used method by industry to produce lactic acid. About 90% of the total worldwide production of lactic acid is made through bacterial fermentation and the rest are produced through the hydrolysis of lactonitrile (Lasprilla et al., 2012; Savioli Lopes et al., 2012). The fermentation process can be carried out in batch, fed-batch, and continuous fermentation. Batch fermentation is the most common fermentation process used in industrial-scale production of lactic acid, because of the ease of sterilization and equipment control. The disadvantage of batch fermentation is that the concentration and productivity of lactic acid produced is relatively low because the final product might inhibit further growth of the bacteria (Borzani et al., 1993). The production of lactic acid through the fermentation process has several advantages over chemical synthesis, i.e. low production costs, low operating temperatures, low energy consumption, and low prices of the substrates used (Abdel-Rahman et al., 2011;

Mayer et al., 2019). The optimum condition for producing lactic acid is pH 6.5, temperature range 30-43°C, and low concentration of oxygen. Lactic acid production through the fermentation process can be considered a green technology because it uses renewable substrates and can also convert CO_2 as a greenhouse gas into lactic acid during the fermentation process (Fachrul Razi, 2006).

The fermentation process to obtain lactic acid takes place either under anaerobic or aerobic conditions based on the type of bacteria used (Matsumoto and Taguchi, 2010; Savioli Lopes et al., 2012). Some lactic acid-producing bacteria are *Lactobacillus amylophilus*, *Lactobacillus bavaricus*, *Lactobacillus casei*, *Lactobacillus maltaromicus*, *Lactobacillus salivarius*, *Lactobacillus delbrueckii*, *Lactobacillus jensenii,* and *Lactobacillus acidophilus* (Lee et al., 2020; Lin et al., 2020). *Lactobacillus sp.* is a bacterium that plays a very important role in producing lactic acid because these bacteria produce the enzyme, α-amylase, which can convert glucose in the substrate into lactic acid (Febriningrum, 2013). Sources of microbial carbon in producing lactic acid can either be: a) pure sugars in the form of glucose, sucrose, lactose; or b) sugar derived from molasses, whey, bagasse, cassava; or c) flour from sweet potatoes, wheat, and barley (Li et al., 2020).

Although the prospect of lactic acid as a bioplastic raw material is very promising, enormous studies are still needed to reduce the cost of PLA production in order to compete with conventional petrochemical-based plastics. It is estimated that the cost of producing lactic acid must be lowered by up to 50% in order for the PLA to be able to compete in the market (Okano et al., 2010; Wee et al., 2004). Several aspects of reducing the cost of lactic acid production have been studied extensively, ranging from raw materials used, selection of microorganisms, substrates, production methods, applications, and others.

In the process of lactic acid fermentation, cost of the substrate consumes as much as 40-70% of the total cost (Rawoof et al., 2021). Thus, the selection of raw materials greatly affects the cost production. The use of polysaccharides such as starch and cellulose require a complex process i.e., delignification process to remove lignin from cellulose, saccharification to break carbohydrate bonds into glucose, and fermentation to produce lactic acid. Pretreatment process in some cases of lactic acid production had increased the production costs (López-Gómez et al., 2019; Rawoof et al., 2021; Wang et al., 2015). One of which as has been reported by (Rahmayetty, D.R Barleany, A. Irawan, 2015) that the synthesis of lactic acid from empty bunches of palm oil (EFB) through the method of simultaneous fermentation saccharification resulted in a low concentration of lactic acid. Although the price of EFB is cheap, the cost for the pretreatment stage resulting in an increase of the entire production cost. The non-optimal EFB delignification process is an obstacle in the fermentation process.

A very simple process to produce lactic acid that requires only one stage of fermentation is the fermentation of glucose. Hence, the use of pure glucose and lactose substrates is not recommended because they are very expensive (Paes et al., 2019). Raw materials that are cheap and can be developed to produce lactic acid are industrial by-product that contain glucose, such as the waste from sugar industry in the form of molasses (Nandasana and Kumar, 2008).

In general, to produce 1 ton of raw sugar, 0.38 tons of molasses will be produced. Molasses cannot be disposed of directly because it is large in quantity with a high content of complex organic compounds and viscosity. Molasses contains 40–60% sugar (w/w) and the water content is below 20% (w/w) (Zhang et al., 2021). Molasses is a substrate that has the potential to be fermented into lactic acid because molasses contains high sugars.

The success of the fermentation process is greatly influenced by several factors such as the type of substrate, pH of the medium, and bacterial concentration. Wee et al. (2004) reported that the use of molasses substrates for lactic acid synthesis by batch fermentation using *Enterococcus faecalis* resulted in a maximum of lactic acid concentration as much as 134.9 g/L. Mahato et al. (2021) conducted a study to optimize the production of lactic acid in a fermentation medium consisting of MRS agar media, MRS broth, and agar with various addition of molasses ranging from 1% to 9% (v / v). Optimum conditions obtained at the addition of molasses as much as 7% (v/v), incubation time of 144 h, and effective agitation speed of 175. Mahato et al. (2021) only used a small amount of molasses as a substrate that was given into a fermentation medium and *Lactobacillus acidophilus* was not through a breeding and adapting process beforehand. The use of small amount of molasses will not solve the environmental problem regarding accumulation of high quantity and undegradable of molasses as the waste in sugar industry. Therefore, it is

necessary to develop research to accommodate a large amount of molasses so that it would no longer be the environmental issue. In this study, molasses fermentation is done without the addition of specific nutrients in the fermentation media, and the *Lactobacillus acidophilus* will be inoculated directly in molasses media. The adaption of bacteria before the fermentation process started is necessary so that the lag phase can be achieved faster in the bioreactor. This study aims to obtain the value of the cell concentration and growth kinetics of *Lactobacillus acidophilus* during the fermentation process that gives the highest concentration of lactic acid.

2. Methodology

2.1. Materials

Molasses was obtained from a sugar factory located in Central Java. *Lactobacillus acidophilus* was obtained from the Microbiology Laboratory of the Bandung Institute of Technology. Medium MRS, NaOH, H_2SO_4, NaOH, and MRS were obtained from Merck, Indonesia.

2.2. Bioreactor Design

The bioreactor used in this study was an anaerobic bioreactor with a volume of 1000 ml. The top of the bioreactor has a rubber cover equipped with a goose-neck flask, a sampling tube and a pipe for conveying N_2 gas. The bioreactor used in this study is shown in Figure 1.

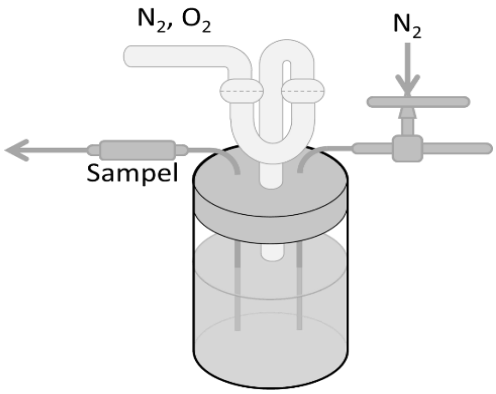


Figure 1. Anaerobic Bioreactor

2.3. *Lactobacillus acidophilus* inoculation

The methodology used in this stage is proposed by Sebayang, (2006) with some modification. At the inoculation stage, 3.7 grams of MRS nutrients were dissolved in 100 mL of distilled water, heated at 100°C, then stirred using a magnetic stirrer at a speed of 150 rpm, continued with sterilization in an autoclave for 15 minutes. The sterilized MRS liquid was then poured into a petri dish that has been previously sterilized. Next, the culture medium was cooled in a closed petri dish until the agar formed. Afterwards, *Lactobacillus acidophilus* were inoculated by applying two oses of the bacteria in the surface of MRS medium in the zigzag pattern and then incubated for 48 hours at room temperature. Bacterial growth is characterized by the appearance of white spots on the surface of the culture medium.

2.4. Molasses pretreatment

Molasses used in this study contained 50.68% total sugar and 32% reducing sugar. Molasses is diluted with the addition of distilled water to a total sugar content of 12-15%.

2.5. Preparation of *Lactobacillus acidophilus* starter

The culturing stage of *Lactobacillus acidophilus* in molasses substrate started by sterilized 500 mL molasses that stored in an Erlenmeyer. Then 25 ml of MRS (1.6 g/100 ml) was added to the starter medium as a nutritional compound. NaOH solution was added until the pH of the medium was 6.0. Afterwards, *Lactobacillus acidophilus* cultures were placed into the starter medium and incubated for 48 hours.

2.6. Molasses Fermentation

The procedure used in this stage was based on previous research Sun et al. (2019) and Wee et al. (2004), with some modification. The fermentation of molasses started when 500 ml of molasses were stored in the bioreactor. The fermentation medium was then sterilized at 120°C for 15 minutes in an autoclave. After the fermentation medium is cooled to room temperature, the starter with various volume; 1, 3, 5 and 10% (v/v), were added. The pH of the culture was maintained at 6.5 by adding 10 M of NaOH. Momentarily flow of nitrogen gas (N_2) into the bioreactor is done to expel oxygen in the bioreactor. The temperature of the bioreactor was kept constant at 35° and kept stirring at 200 rpm so that the fermentation broth was completely mixed. Fermentation was carried out for 72 hours and samples were taken every 8 hours.

2.7. Lactic acid concentration analysis

The concentration of lactic acid produced in the fermentation process was analysed using

UV-vis spectrophotometry at 284 nm and compared with a standard solution of pure lactic acid.

2.8. Analysis of *Lactobacillus acidophilus* concentration

Bacterial concentration was measured by gravimetric method. Bacterial growth is identical to the addition of Mixed Liquor Suspended Solid (MLSS). Bacterial concentration test was carried out by filtering a number of samples using 0.45 m filter paper. The filter results were dried at a temperature of 103-105^{o}C for one hour to obtain a constant weight. The filtered sample was weighed and the MLSS concentration is calculated by equation 1.

$$\text{Concentration of cells } \left(\frac{\text{mg}}{\text{L}}\right) = \frac{(\text{A} - \text{B})\text{x}1000}{\text{ml sample}} \qquad (1)$$

where A is weight of filter paper containing residue and B is empty filter paper weight.

2.9 Determination of Cell Growth Kinetics using Logistic Equation model

The growth of *Lactobacillus acidophilus* in the fermentation medium can be determined using a logistic growth model. In this logistic growth equation, it is assumed that there is no time delay in the growth process of microorganisms, whether the microorganisms will continue to increase (never decrease) or will continue to decrease (never increase) (Panigoro, 2015). The specific growth rate was calculated by equation 2 and maximum net specific growth rate (μ_{net}) was calculated by equation 3 (Shuler and Kargi, 1991).

$$k = \frac{1}{\bar{X}}\frac{\Delta X}{\Delta t} + \left[1 - \frac{\bar{X}}{X_\infty}\right] \qquad (2)$$

where k is the carrying-capacity coefficient, Δt is time period (t_n-t_{n-1}), $\bar{X}$ is average biomass concentration during Δt, ΔX is the difference in the biomass concentration, and X_∞ is the maximum cell mass.

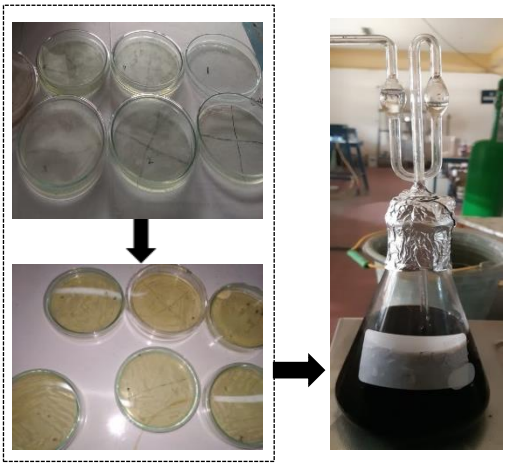
$$\mu_{net} = \frac{lnX_2 - lnX_1}{t_2 - t_1} \qquad (3)$$

where μ_{net} is the maximum net specific growth rate (h^{-1}), X_1 is the initial cell mass concentration of the exponential growth phase (mg/L), and X_2 is the final cell mass concentration of the exponential growth phase (mg/L).

3. Results and Discussion

3.1. Inoculation and culturing of *Lactobacillus acidophilus*

Lactobacillus acidophilus was inoculated in MRS media using the quadrant streak plate method. *Lactobacillus acidophilus* inoculum was then cultured in molasses medium as shown in Figures 2(a) and (b).



(a) (b)

Figure 2. (a) inoculum of *Lactobacillus acidophilus*; (b) starter of *Lactobacillus acidophilus*

Figure 2(a) shows *Lactobacillus acidophilus* inoculation for 24 hours on MRS medium. The formation of bacterial colonies in *Lactobacillus acidophilus* inoculation was characterized by the color change in the MRS medium. Figure 2(b) displays the process of *Lactobacillus acidophilus* in molasses as a starter. The starter bacteria are allowed to breed and adapt in the substrates, that have the same nutrients composition with the fermentation medium. Bacterial cultures were then mixed evenly in molasses medium. During the formation of the starter, the concentration of bacteria gradually increased until the end of the process, as can be observed in Table 1. The increase in the concentration of cells in the starter indicated that *Lactobacillus acidophilus* grew well on molasses medium.

Table 1. Concentration of *Lactobacillus acidophilus* during the starter stage

Time (hour)	concentration of bacteria (mg/L)	pH Medium
0	148.2	6.0
8	163.5	5.9
16	226.8	5.7
24	388.3	5.6
32	424.7	5.3
48	535.5	5.1

The adaptation phase (lag phase) of the bacteria in the new environment, that occurs immediately after inoculation, can be achieved less than 8 hours. *Lactobacillus acidophilus* is a species of bacterium that easily adapts to the surrounding environment, so it does not take a long time to reach a fast growth phase (Horackova et al., 2020).

3.2. Molasses Fermentation

In the fermentation process, increasing the concentration of *Lactobacillus acidophilus* (1; 3; 5; and 10%(v/v)) in the molasses medium decreased the pH of the mixture. The decreasing of pH in the mixture can be observed in Figure 3. At the eighth hour it was seen that the pH of the mixture did not significantly decrease. A significant decrease in pH occurred after 24 hours of fermentation. This is caused by the accumulation of lactic acid and the amount of natural buffer in the medium that can no longer support the formation of acid, thus it continued to decrease the pH until the end of the fermentation process. *Lactobacillus acidophilus* starter can work in a fairly wide range of pH, 3.5-6.0, with the optimum pH range 5-5.8. The lower the pH, the more lactic acid formed in the bioreactor. A significant decrease in pH 5.0, occurred at a 10% concentration of *Lactobacillus acidophilus* at the end of the fermentation process (72 hours). Tay and Yang, (2002) reported that the formation of lactic acid, ethanol, and fumaric acid reduced the pH of the medium to 4.0. Cell concentration changes during the fermentation process are shown in Figure 4.

During the fermentation process it was seen that the cell concentration changed over time. In Figure 4, it can be seen that the cells experienced a lag phase, an exponential growth phase, and a stationary phase. The lag phase occurs at the beginning of the fermentation process. With the addition of starter 1 and 3% (v/v) it was seen that the lag phase occurred in the first 16 hours of the fermentation process, after which the cells experienced a rapid growth phase until the end of the fermentation process. In both variations, cells do not experience a stationary phase and a death phase. This indicates that the cell still has sufficient substrate for its growth. At 5% starter volume variation, it was seen that the cells only experienced an exponential phase. This is indicated by the increasing concentration of cells in the fermentation medium.

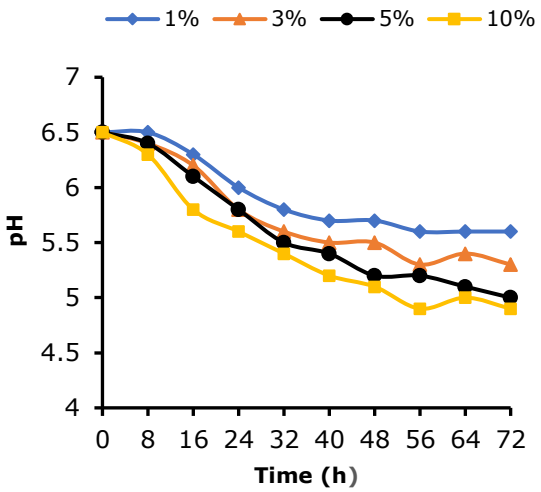


Figure 3. pH in fermentation process

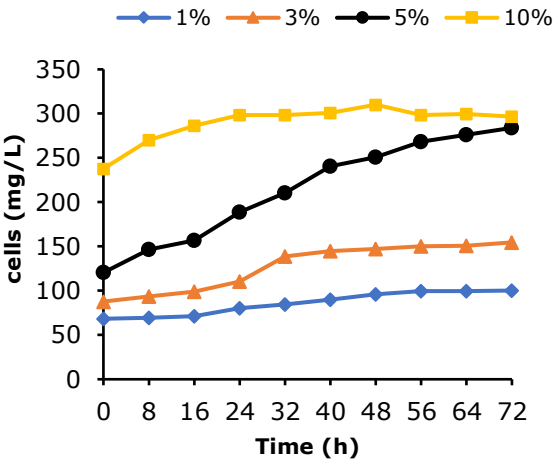


Figure 4. Cells concentration in fermentation process

Sufficient amount of substrate and suitable environmental conditions cause cell multiplication to take place optimally (Rolfe et al., 2012). With the addition of 10% starter volume, it was seen that the cells underwent an exponential phase for 32 hours of the fermentation process. The stationary phase occurred at 32-40 hours of the fermentation process, followed by a dead phase. The stationary phase is indicated by the number of cells that does not significantly change, which is caused by a balance between the rate of growth and the cell death. The death phase is indicated by a decrease in cell concentration. This is caused by an imbalance in the rate of cell growth and death. Cell death occurs because the substrate begins to decrease and the products that are toxic to microorganisms starts to accumulate (Rolfe et al., 2012).

The concentration of lactic acid production during fermentation process can be observed in Figure 5.

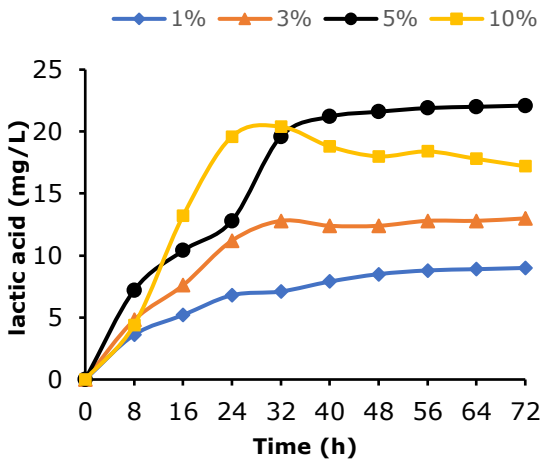


Figure 5. Lactic acid production during the fermentation process

The carbon source in the form of sugar in the medium during the fermentation process will be converted into lactic acid and new cells. The graphic at Figure 5 shows that the concentration of lactic acid increased during the fermentation process. At the addition of 10% starter, the concentration of lactic acid increased significantly after 8 hours of fermentation. This result shows the ability of the bacteria to optimally utilize the substrate to convert molasses into lactic acid. At 32 hours of fermentation, there was a decrease in the concentration of lactic acid. The decrease in the concentration of lactic acid might occur as an effect of further reaction that converts lactic acid into other compounds that might inhibit the cell growth in the fermentation media (Mirdamadi et al., 2002). This product inhibition can be reduced by providing more nutrients needed by microorganisms (Bulut et al., 2004). The highest concentration of lactic acid, produced during 72 hours of fermentation, was 23.1 ppm. With the addition of 5% starter volume, the concentration of lactic acid increased rapidly during 24-32 hours of fermentation. The graphic also displays the result of starter 1 and 3% addition that shows a significant increase of the concentration of lactic acid between 0-32 hours, but tends to be constant between 32 to 72 hours of the fermentation process.

These results have similarities with those reported by Idris and Suzana, (2006) and Mussatto et al. (2008) that these bacteria can effectively convert molasses into lactic acid in the first 12 hours of the fermentationprocess.The decrease in the activity of microorganisms might be an effect of the uncontrolled of pH system. The pH decreases might occur due to the conversion of glucose into lactic acid and other acids. pH is an important factor in the growth and activity of microorganisms, thus the optimum pH for growth and production of lactic acid has to be maintained in the range of 5.0 - 7.0 (Hofvendahl and Hahn-Hägerdal, 2000). The optimum conditions for *Lactobacillus acidophilus* to grow and effectively convert molasses into lactic acids are pH 6.5 and temperature 37°C (Tomás et al., 2003). Some other fermentation of molasses research to yield lactic acid using batched process can be observed in Table 2.

Tabel 2. Comparison of batch fermentation of molasses to lactic acid

Organism	Lactic acid, mg/L	Fer. Time, h	Ref
Lactobacillus delbrueckii	1.66. 10^5	40	Dumbrepatil et al., 2008
Enterococcus faecalis RKY1	6.51. 10^4	30	Wee et al., 2004
Enterococcus hirae ds 10	8.60. 10^2	36	Abdel-Rahman et al., 2020
Lactobacillus acidophilus	12.38	72	Mahato et al., 2021
Lactobacillus acidophilus	23.1	72	This study

3.3. Cell Growth Kinetics

The research variation with 5% starter volume was chosen for the calculation of the specific growth rate because it produced the highest concentration of lactic acid, 23.1 mg/L, with the value of X∞=283.9 mg/L, at the fermentation time (t) = 72 hours, which means that the cell growth was almost complete at 72 hours. This logistic growth curve can be observed in Figure 6.

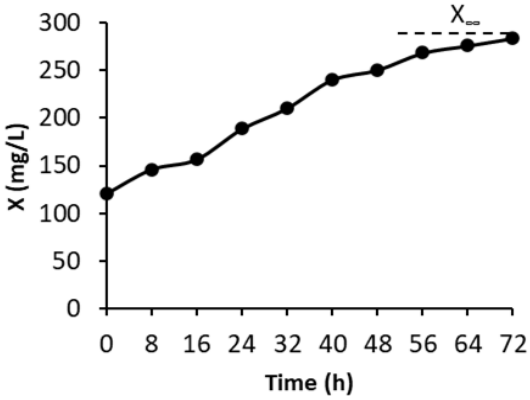


Figure 6. Logistic growth curve of *Lactobacillus acidophilus*

The cell specific growth rate was determined using equation 2 and the results are presented in Table 3.

Table 3. Cell growth kinetics with of 5% *Lactobacillus acidophilus*

$\Delta t\ (h)$	$\bar{X}$ (mg/L)	$\frac{1}{\bar{X}}\frac{\Delta X}{\Delta t}\ (\mathrm{h}^{-1})$	$\left[1-\frac{\bar{X}}{X_\infty}\right]$	k (h^{-1})
8	122.0	0.00317	0.5701	0.5733
8	136.2	0.02304	0.5204	0.5435
8	167.7	0.02826	0.4095	0.4378
8	204.6	0.02194	0.2796	0.3015
8	238.2	0.01643	0.1612	0.1776
8	264.6	0.01020	0.0681	0.0783
8	278.9	0.00314	0.0177	0.0208
8	282.4	0.00004	0.0052	0.0052
8	283.2	0.00063	0.0025	0.00313

The mean value of the cell growth kinetics is k = 0.2379 h^{-1}. The maximum net specific growth rate was determined when *Lactobacillus acidophilus* is in exponential phase. The maximum net specific growth rate (μ_{net}) is calculated using equation (3).

$$\mu_{net} = \frac{ln282.5 - \ln 167.7}{56 - 16} = 0{,}0160\ h^{-1}$$

The comparison of k and μ_{net} for the starter volume variation is shown in Table 4.

Table 4. Growth kinetics parameters

Starter Volume (%)	X_0 (mg/L)	k (h^{-1})	$\mu_{net}(h^{-1})$
1	68.32	0.3926	0.0046
3	87.76	0.2799	0.0092
5	120.60	0.2379	0.0160
10	237.81	-0.0225	0.0032

The calculation of each variation of the initial concentration of cells resulted to different value of k. The value of k describes the growth capacity of a population (k value > 0). The greater value of k indicates the greater carrying capacity of cell growth. The value of k close to 0 indicates that the carrying capacity of the substrate is limited, which means that the growth of microorganisms is approaching the stationary phase (Timuneno et al., 2008).

The data in Table 4 presented the result that the greater concentration of cells added at the beginning of the fermentation process, the smaller value of the carrying capacity coefficient (k). This indicates that cell growth is heading to the stationary phase which also supported by the data of the increasing in the maximum net specific growth rate (μ_{net}). The addition of 10% starter volume resulted in a negative k value. This indicates that the cells experienced a death phase during the fermentation process. This is also supported by the value of μ_{net} which is smaller than the other variations. Thus, this data cannot be used to calculate cell growth kinetics. The logistic model assumes that at any given time of the population will approach the equilibrium point. At this point the number of births and deaths is considered the same, so the graph will be at the steady state (Timuneno et al., 2008).

4. Conclusion

The highest concentration of lactic acid produced in batch molasses fermentation was 23.1 mg/L, resulted from the addition of 5% starter volume and 72 hours fermentation time. By using the logistic model, it can be concluded that the value of growth capacity will approach the stationary phase faster if more of the starter added in the fermentation media. The addition of excess starter to the fermentation media causes a faster death phase because the amount of substrate is limited and the accumulation of lactic acid will also inhibit the cell growth. The value of k that is close to equilibrium and the highest net is obtained with the addition of 5% starter with the values of k = 0.2379 h^{-1} and μ_{net} = 0.0160 h^{-1} respectively.

References

Abdel-Rahman, M.A., Hassan, S.E.D., El-Din, M.N., Azab, M.S., El-Belely, E.F., Alrefaey, H.M.A., Elsakhawy, T., 2020. One-factor-at-a-time and response surface statistical designs for improved lactic acid production from beet molasses by Enterococcus hirae ds10. SN Appl. Sci. 2. https://doi.org/10.1007/s42452-020-2351-x

Abdel-Rahman, M.A., Tashiro, Y., Sonomoto, K., 2011. Lactic acid production from lignocellulose-derived sugars using lactic acid bacteria: Overview and limits. J. Biotechnol.https://doi.org/10.1016/j.jbiotec.2011.06.017

Borzani, W., Sanchez Podlech, P.A., Luna, M.F., Jerke, P.R., Stein, M.A.C.F., 1993. Kinetics of semicontinuous microbial transformation of whey by Lactobacillus bulgaricus varying the initial concentration of yeast autolysate. J. Biotechnol. 31, 61–66. https://doi.org/10.1016/0168-1656(93)90136-B

Bulut, S., Elibol, M., Ozer, D., 2004. Effect of different carbon sources on L(+) -lactic acid production by Rhizopus oryzae. Biochem. Eng. J. 21, 33–37. https://doi.org/10.1016/j.bej.2004.04.006

Dumbrepatil, A., Adsul, M., Chaudhari, S., Khire, J., Gokhale, D., 2008. Utilization of molasses sugar for lactic acid production by Lactobacillus delbrueckii subsp. delbrueckii mutant Uc-3 in batch fermentation. Appl. Environ. Microbiol. 74, 333–335. https://doi.org/10.1128/AEM.01595-07

Fachrul Razi, and S.. Y., 2006. Batch and Continuous Lactic Acid Production from Cassava by Streptococcus bovis. Rekayasa Kim. dan Lingkung. 5, 17–20.

Febriningrum, P.N., 2013. Pengaruh Konsentrasi Substrat Kulit Nanas dan Kecepatan Pengadukan terhadap Pertumbuhan Lactobacillus plantarum untuk Produksi Asam Laktat. J. Rekayasa Kimia Lingkungan, 9, 144. https://doi.org/10.23955/rkl.v9i3.784

Hofvendahl, K., Hahn-Hägerdal, B., 2000. Factors affecting the fermentative lactic acid production from renewable resources. Enzyme Microb. Technol. 26, 87–107.https://doi.org/10.1016/S0141-0229(99)00155-6

Horackova, S., Vesela, K., Klojdova, I., Bercikova, M., Plockova, M., 2020. Bile salt hydrolase activity, growth characteristics and surface properties in Lactobacillus acidophilus. Eur. Food Res. Technol. 246, 1627–1636. https://doi.org/10.1007/s00217-020-03518-8

Idris, A., Suzana, W., 2006. Effect of sodium alginate concentration, bead diameter, initial pH and temperature on lactic acid production from pineapple waste using immobilized Lactobacillus delbrueckii. Process Biochem. 41, 1117–1123. https://doi.org/10.1016/j.procbio.2005.12.002

Komesu, A., de Oliveira, J.A.R., da Silva Martins, L.H., Maciel, M.R.W., Filho, R.M., 2017. Lactic Acid Production to Purification: A Review. BioResources 12, 4364–4383. https://doi.org/10.15376/BIORES.12.2.KOMESU

Lasprilla, A.J.R., Martinez, G.A.R., Lunelli, B.H., Jardini, A.L., Filho, R.M., 2012. Poly-lactic acid synthesis for application in biomedical devices - A review. Biotechnol. Adv. https://doi.org/10.1016/j.biotechadv.2011.06.019

Lee, J.M., Jang, W.J., Lee, E.W., Kong, I.S., 2020. β-glucooligosaccharides derived from barley β-glucan promote growth of lactic acid bacteria and enhance nisin Z secretion by Lactococcus lactis. LWT 122. https://doi.org/10.1016/j.lwt.2020.109014

Li, G., Zhao, M., Xu, F., Yang, B., Li, X., Meng, X., Teng, L., Sun, F., Li, Y., 2020. Synthesis and Biological Application of Polylactic Acid. Molecules. https://doi.org/10.3390/molecules25215023

Lin, H.T.V., Huang, M.Y., Kao, T.Y., Lu, W.J., Lin, H.J., Pan, C.L., 2020. Production of lactic acid from seaweed hydrolysates via lactic acid bacteria fermentation. Fermentation 6. https://doi.org/10.3390/FERMENTATION6010037

López-Gómez, J.P., Alexandri, M., Schneider, R., Venus, J., 2019. A review on the current developments in continuous lactic acid fermentations and case studies utilising inexpensive raw materials. Process Biochem. https://doi.org/10.1016/j.procbio.2018.12.012

Mahato, A.K., Kumari, L., Singh, R.S., Alam, M.T., 2021. Fermentative study on optimization of lactic acid production from cane sugar by Lactobacillus spp. Eur. J. Mol. Clin. Med. 8, 712–723.

Matsumoto, K., Taguchi, S., 2010. Enzymatic and whole-cell synthesis of lactate-containing polyesters: Toward the complete biological production of polylactate. Appl. Microbiol. Biotechnol. https://doi.org/10.1007/s00253-009-2374-0

Mayer, F., Bhandari, R., Gäth, S., 2019. Critical review on life cycle assessment of conventional and innovative waste-to-energy technologies. Sci. Total Environ. https://doi.org/10.1016/j.scitotenv.2019.03.449

Mirdamadi, S., Sadeghi, H., Sharafi, N., Fallahpour, M., Mohseni, F.A., Bakhtiari, M.R., 2002. Comparison of lactic acid isomers produced by fungal and bacterial strains. Iran. Biomed. J. 6, 69–75.

Mussatto, S.I., Fernandes, M., Mancilha, I.M., Roberto, I.C., 2008. Effects of medium supplementation and pH control on lactic acid production from brewer's spent grain. Biochem. Eng. J. 40, 437–444. https://doi.org/10.1016/j.bej.2008.01.013

Nandasana, A.D., Kumar, S., 2008. Kinetic modeling of lactic acid production from molasses using Enterococcus faecalis RKY1. Biochem. Eng. J. 38, 277–284. https://doi.org/10.1016/j.bej.2007.07.014

Okano, K., Tanaka, T., Ogino, C., Fukuda, H., Kondo, A., 2010. Biotechnological production of enantiomeric pure lactic acid from renewable resources: Recent achievements, perspectives, and limits. Appl. Microbiol. Biotechnol. https://doi.org/10.1007/s00253-009-2280-5

Paes, L.A.B., Bezerra, B.S., Deus, R.M., Jugend, D., Battistelle, R.A.G., 2019. Organic solid waste management in a circular economy perspective – A systematic review and SWOT analysis. J. Clean. Prod. 239. https://doi.org/10.1016/j.jclepro.2019.118086

Panigoro, H.S., 2015. Model Pertumbuhan Logistik: Modifikasi pada Daya Dukung dengan Pemanenan Proposional terhadap Populasi, in: Konferensi Nasional Pendidikan Matematika-VI. pp. 267–273.

Rahmayetty, D.R Barleany, A. Irawan, E.S., 2015. Sintesis Asam Laktat Berbahan Baku Tandan Kosong Kelapa Sawit Menggunakan Trichoderma Reseei dan Lactobacillus acidiphillus, in: Seminar Nasional Sains Dan Teknologi.

Rawoof, S.A.A., Kumar, P.S., Vo, D.V.N., Devaraj, K., Mani, Y., Devaraj, T., Subramanian, S., 2021. Production of optically pure lactic acid by microbial fermentation: a review. Environ. Chem. Lett. https://doi.org/10.1007/s10311-020-01083-w

Rolfe, M.D., Rice, C.J., Lucchini, S., Pin, C., Thompson, A., Cameron, A.D.S., Alston, M., Stringer, M.F., Betts, R.P., Baranyi, J., Peck, M.W., Hinton, J.C.D., 2012. Lag phase is a distinct growth phase that prepares bacteria for exponential growth and involves transient metal accumulation. J. Bacteriol. 194, 686–701. https://doi.org/10.1128/JB.06112-11

Savioli Lopes, M., Jardini, A.L., Maciel Filho, R., 2012. Poly (lactic acid) production for tissue engineering applications, in: Procedia Engineering. pp. 1402–1413. https://doi.org/10.1016/j.proeng.2012.07.534

Sebayang, F., 2006. Pembuatan Etanol dari Molase Secara Fermentasi Menggunakan Sel Saccharomyces cerevisiae yang Terimobilisasi pada Kalsium Alginat. J. Teknol. Proses Media Publ. Karya Ilm. Tek. Kim. 5, 75–80.

Shuler, M.L., Kargi, F., 1991. Bioprocess Engineering Basic Concepts Second Edition. Adv. Chem. Eng. 535.

Sun, Y., Xu, Z., Zheng, Y., Zhou, J., Xiu, Z., 2019. Efficient production of lactic acid from sugarcane molasses by a newly microbial consortium CEE-DL15. Process Biochem. 81, 132–138. https://doi.org/10.1016/j.procbio.2019.03.022

Tay, A., Yang, S.T., 2002. Production of L(+)-lactic acid from glucose and starch by immobilized cells of Rhizopus oryzae in a rotating fibrous bed bioreactor. Biotechnol. Bioeng. 80, 1–12. https://doi.org/10.1002/bit.10340

Timuneno, H.M., Utomo, R.H.S., Widowati, 2008. MODEL PERTUMBUHAN LOGISTIK DENGAN WAKTU TUNDA. J. Mat. 11 (1), 43–51.

Tomás, M.S.J., Ocaña, V.S., Wiese, B., Nader-Macías, M.E., 2003. Growth and lactic acid production by vaginal Lactobacillus acidophilus CRL 1259, and inhibition of uropathogenic Escherichia coli. J. Med. Microbiol. 52, 1117–1124. https://doi.org/10.1099/jmm.0.05155-0

Wang, Y., Tashiro, Y., Sonomoto, K., 2015. Fermentative production of lactic acid from renewable materials: Recent achievements, prospects, and limits. J.

Biosci. Bioeng. https://doi.org/10.1016/j. jbiosc.2014.06.003

Wee, Y.J., Kim, J.N., Yun, J.S., Ryu, H.W., 2004. Utilization of sugar molasses for economical L(+)-lactic acid production by batch fermentation of Enterococcus faecalis, in: Enzyme and Microbial Technology. pp. 568–573. https://doi.org/10.1016/j.enzmictec.2004.08.008

Zhang, S., Wang, J., Jiang, H., 2021. Microbial production of value-added bioproducts and enzymes from molasses, a by-product of sugar industry. Food Chem. https://doi.org/10.1016/j.foodchem.2020.128860