The effect of cold storage temperatures on respiration rate and physical quality of crownless pineapple (*Ananas comosus* L.)

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Abstract. Pineapple is a horticultural product with high consumption and production. Also, this fruit has a perishable characteristic. It is influenced by physiological processes, which are respiration and transpiration. It needs good handling during the post-harvest process to control the freshness and quality, especially in the storage process. Thus, it is important to know the effect of low storage temperature on the respiration rate and physical quality of pineapple. In this study, a sample conducted triplicate and observed over a period by treating crownless pineapple in cold storage and storage temperature maintain at 7°C and 90% of relative humidity. The physical quality properties are weight loss, water content, shell and flesh colour, acidity, total dissolved solids (TDS), and hardness. The respiration sample conducted a closed method with the plastic jar as a container to observe the changes of O2 and CO2 at 2, 4, 12, 24 hours intervals over 7.5 days of storage. The physical quality properties sample put in a carton box to observe the changes in the quality at 2-3 days intervals over 24 days. The resulted of respiration rate and physical properties changes slowly during storage. Some sample observed chilling injury with the black spot on the shell of the fruit. Overall, the weight loss, water content, and hardness decreased, and the colour changed from green to yellow in half of the fruit during storage.

1. Introduction

Pineapple is a horticultural product with high consumption and production. In 2016, a total of 26.4 million tonnes was marketed. Over the past nine years, the market has grown on average by 3.3% per year. The consumption of pineapples continues to grow, in part due to the rising income and growing population, but also to marketing campaigns focused on healthy food consumption. On the production side, the total volume in 2016 worldwide is 26.67 million tonnes. Costa Rica is undoubtedly number one, with exports total of 3.2 million tonnes or 12% of worldwide production. Next, in the top 3 are Brazil (10%), Philippines (10%), and Indonesia being number nine with 5% [1].

Pineapple is widely distributed on the market in the form of whole fruit with crown and crownless. In addition, fresh-cut fruit is also widely available in fruit shops and markets. Crown pineapple is marketed to provide a selection of fresh fruit with a complete structure while crownless pineapple is marketed to overcome the crown damage to keep it looking attractive to the consumer. There are other products of processed pineapple, such as dried and canned pineapple in various forms (chunks, tidbits, slices, and spears). All of them provide choices for fruit products with different freshness, shapes, and nutritional content.
The post-harvest of pineapple starts from harvesting, collecting, sorting, classifying, packaging, labelling, storing, and distributing [2]. It takes a relatively long time for the storage process, so it needs good handling to prevent quality degradation such as mould, decay, shell pitting, chilling injury, and internal browning [3]. Thus, the storage technology needed for storing fruits in optimal storage conditions [4]. The common technology used in the industry to store their product is a cold storage machine. This machine conditioned the storage room at low temperatures and high humidity treatments.

Quality degradation caused by a physiological process (respiration, transpiration, and ethylene production) reacts chemicals with enzymes in cells [5]. Respiration is the process of breaking down complex substrates such as carbohydrates, proteins, and fats react with O₂ to be a simpler component of CO₂, H₂O, and energy. The respiration rate is higher if the storage temperature is higher than the optimal point. It causes the fruit to decay and decrease in physical quality properties rapidly. Therefore, research needs to be done with the objectives to determine 1) the changes of respiration rate during storage and 2) The changes of physical parameters (weight loss, water content, shell and flesh colour, acidity, TDS and hardness).

2. Methodology
2.1. Experimental Procedures
2.1.1. Samples Preparation
Smooth Cayenne Sunpride fresh pineapple fruit with crowns provided by PT. Great Giant Pineapple, Lampung, Indonesia. Product transported by refrigerated container trucks from the company until arriving for approximately 2 days. The fruits were chosen with a standard size of 3-4 (1.8-2.5 kg) and the maturity level 0-10%. The pineapples had cleaned the dirt manually with hand and duster. The crown leave 3 cm above the fruit. The respiration sample inserted into plastic jars as a container. In the lid portion, the jar is given the seal tape to tightening the closure. At the top of the lid, the jar was given a hole in two places and given a glue seal to ensure not to leak the gas. This hole was used to suction gas samples to be measured in a gas analyser and as a gas return hole that has passed through the gas analyser. The physical quality sample put in a carton box to observe the changes in the quality at 2-3 days intervals over 24 days. One box contains 7 fruits of pineapple and each box put in a wood rack on cold storage machine. After that, it was placed on a shelf in the storage room that has been conditioned. The temperature in the storage room of pineapple maintained at 7°C while the storage humidity maintained at 90% using a thermohygrometer. Each interval time, the sample of fruit pick up to measure the quality properties in laboratories of Food and Post-harvest Engineering, Universitas Gadjah Mada.

2.1.2. Mass and Volume Measurement
Weight of fruit was measured with a digital analytical scale. The volume of fruit determined based on the fruit density by cutting the fruit flesh in a cubic shape with 2 cm size. The height, length, and width of the sample measured using a ruler to calculate the sample volume and furthermore to obtained density. The density of samples used to determine the whole fruit volume. The volume of jars was measured by the water displacement method. The average values of the triplicate weight of fruit and free volume are 2.032 kg and 32.086 litres. The free volume of the jars consisted of the total volume of the jars minus the volume occupied by their content.

2.1.3. Respiration Measurement
The respiration rates of pineapple were measured using a closed system method [6-9]. The pineapple with a crown was placed in polypropylene jars containing air as the initial gas atmosphere. The jars were simply closed with a plastic lid with two valves to flow the gas. The sampling process of the headspace gas concentrations was terminated when the constant rate period reached. Headspace O₂ and CO₂ gas concentrations in each jar were monitored using a Gasin-DH JD200 Tianjin gas analyser. Gas samples were taken by insert a needle through to the first valve of the jars. Gas readings were
taken continuously until constant data were displayed on the screen. To avoid modifications in the headspace gas composition and pressure due to gas sampling, a return needle was also attached to the second valve. Typically, the samples were taken every 2 hours for the first 12 hours, then 4 hours for 24 hours, then 12 hours for 5 days, and finally every day. Changes in O\textsubscript{2} and CO\textsubscript{2} concentrations were used to estimate the respiration rate and modelling the respiration at another various temperature. Experiments were conducted at 7 °C, and the samples were conducted in triplicate each storage temperature.

2.1.4. Physical Properties Measurement
Measurements of physical properties have been carried out were weight loss, moisture content, flesh and shell fruit colour, acidity (pH), total dissolved solids (TDS), and hardness. For weight loss measurements carried out by the method of scaling the first time and final time of fruit mass after a certain time interval. For measurement of water content carried out by the method of thermogravimeter. At first, a sample of cubic 2 cm size was prepared, three replications were made and scaled, after that, it was put into the oven for 24 hours, and the final sample was scaled again. The difference in mass is the water content in the sample. Measurement of the color of the flesh and shell of the fruit is done by shooting with colour meter 9 times in different places on the shell of fruit and 6 times in different places on the flesh of the fruit. Measurement of acidity and TDS was done by pounding the sample until the water comes out on a small cup of 10 ml volume and then tested with a refractometer and a pH meter where the results of the reading were the results of the data without further mathematical analysis. The hardness measurement was done by preparing 3 cm x 3 cm x 3 cm sample blocks with 3 replications and then tested by Brookfield Texture Analyser.

2.2. Mathematical Procedures
2.2.1. Respiration Rate
Respiration rate during storage time was calculated through O\textsubscript{2} consumption and CO\textsubscript{2} production measurements. O\textsubscript{2} consumption and CO\textsubscript{2} production rates were determined as [10-11].

\[
RR = \frac{dy}{dt} \times \frac{Vf}{M} 
\]

, where \(RR\) is the tissue respiration rate in O\textsubscript{2} consumption and CO\textsubscript{2} production (ml kg\(^{-1}\) h\(^{-1}\)), \(dy\) is the changes of gas concentration (%) during the time interval as \(dt\) (h), \(Vf\) is the headspace volume (mL), and \(M\) is the sample mass (kg).

2.2.2. Respiration Rate Modelling
The respiration rate of fruit tissues refers to a global process that encompasses the diffusion of gases through the tissues as well as respiration at the cellular level [12]. Several models describe the relationship between gas concentrations and the respiration rate of fruit, although enzymatic models or Michaelis-Menten analysis have been chosen in many cases [13-14]. An uncompetitive inhibition model [15-18] was also fitted to the data to study the possible effect of CO\textsubscript{2} concentration on O\textsubscript{2} consumption (eqs. 2.2 and 2.3).

\[
\frac{dy_{O2}}{dt} = \frac{Vmo. y_{O2}}{Kmo + \left(1 + \frac{V_{CO2}}{Kto}\right)y_{O2}} \times \frac{M}{Vf} \tag{2.2}
\]

\[
\frac{dy_{CO2}}{dt} = \frac{Vmc. y_{O2}}{Kmc + \left(1 + \frac{V_{CO2}}{Ktc}\right)y_{O2}} \times \frac{M}{Vf} \tag{2.3}
\]
Where $V_m$ is the maximum respiration rate, $K_m$ is the oxygen concentration at half of $V_m$, $K_i$ is the dissociation constant, and $o$ as $O_2$ and $c$ as $CO_2$. The eqs. 2.3 was used to estimate the model parameters by non-stiff Ordinary Differential Equations (non-stiff ODEs) 45 and fminsearch algorithm solver type using Matlab 2013a software. The solver function implements a Runge-Kutta method with a variable time step for efficient computation. The determination coefficient ($R^2$) and the sum-square of error (SSE) were calculated to evaluate the accuracy of the model. In general, the lower the SSE, the better the agreement between the experimental and predicted data [19].

2.2.3. Weight Losses
The changes in weight loss obtained from the initial weight measurement minus the final weight and divided by initial weight and multiply 100% in Equation 2.2 below. Where WL is weight loss (%), $m_0$ is an initial mass (g), and $m_f$ is the final mass (g).

$$WL = \frac{m_0 - m_f}{m_0} \times 100\%$$

(2.4)

2.2.4. Water Content
The water content used in this study is the dry basis water content ($WC_{db}$). Dry basis water content is the percentage of the water content of an ingredient expressed based on the wet weight per dry weight. Value of $WC_{db}$ can be seen in Equation 2.3.

$$WC_{db} = \frac{m_{c+s} - m_{c+so}}{m_{c+so} - m_c} \times 100\%$$

(2.5)

Where $WC_{db}$ is water content dry basis (%), $m_{c+s}$ is the weight of the cup and sample before putting in oven (g), $m_{c+so}$ is the weight of cup and sample after drying in the oven (g), and $m_c$ is the cup weight (g).

2.2.5. Shell and Flesh Colour
Colour changes were analysed using the values of lightness, hue, and chroma that were obtained from the colourmeter. Equations 2.4, 2.5, and 2.6 are used to find the value of hue and chroma. Where $L$ is lightness (%), $H$ is hue ($^\circ$), $C$ is chroma (%), $a$ is redness-green value, and $b$ is yellowness-blueness value.

$$L = L(\%)$$

(2.6)

$$H = \arctan\left(\frac{b}{a}\right)$$

(2.7)

$$C = \sqrt{a^2 + b^2}$$

(2.8)

2.2.6. Hardness
The hardness values were obtained from calculations based on Equation 2.7, where the value of $F$ is the compressive force (kgf), and $A$ (cm$^2$) is the surface area of the probe used in the texture analyzer.

$$P = \frac{F}{A}$$

(2.9)

3. Results and Discussions
3.1. Respiration Rate
After being harvested, the fruit is still doing respiration. It is proven by knowing the changes of $O_2$ and $CO_2$ concentration inside the jars of pineapple. Changes in concentration under closed-system conditions make the $O_2$ decreases, and the $CO_2$ increases continuously to a certain period. Figure 1a showed the change in $O_2$ and $CO_2$ during storage, and figure 1b showed the respiration rate during storage at cold temperature. The results showed that changes in $O_2$ decreased while changes in $CO_2$ increased during storage times. Changes in $O_2$ and $CO_2$ gas observed at a low rate.
The lowest respiration rate resulted at 7°C with the value is 2-4 mgCO₂ kg⁻¹h⁻¹, while the higher respiration rate resulted at a higher temperature. The results show that at the low storage temperature effect on the low respiration rate. It caused by the cellular of the tissues process respiration slowly. Thus, the consumption rate of O₂ and the production of CO₂ run slower too. The average respiration rate of O₂ consumed is 1.72 ml kg⁻¹h⁻¹ and CO₂ produced is 7.22 ml kg⁻¹h⁻¹ [3].

Figure 1. Changes of O₂ (○) and CO₂ (▲) (a) gas concentrations inside closed pineapple jars and (b) respiration rate until the constant period

3.2. Respiration Rate Modelling

Figure 2 showed changes in O₂ and CO₂ based on Michaelis-Menten modelling. Table 1 showed Michaelis-Menten constant at 7 °C. The results showed that the modelling has a good fit for the observation with determination coefficient 0.85 for CO₂ and 0.91 for O₂. Although there was a negative constant showed in Kmo parameter, but another research resulted in the same thing, for example, research of tomatoes [20] and blueberry [21]. It is caused by negative O₂ and CO₂ observation data. Thus, the best result with good SSE and determination coefficient is negative constant.

Figure 2. (○) O₂ consumption and (△) CO₂ produce fit in Michaelis–Menten model (continuous lines).

Table. 1 Michaelis-Menten constant at 7°C

|      | value  |
|------|--------|
| Vmo  | 10.796 |
| Vmc  | 16.344 |
| Kmo  | -2.463 |
| Kmc  | 3.762  |
| Kio  | 0.002  |
| Kic  | 0.007  |
3.3. Weight Losses

Weight loss during storage shows a relatively constant rate with an average of 0.2%/day. This indicates that very little change in weight loss is due to the influence of cold temperatures. Higher storage temperatures will increase the respiration rate of the fruit, and lower temperatures will inhibit the fruit respiration rate. The rate of respiration affects the rate of metabolism and the storage life of pineapple fruit. The higher respiration rate it makes fruit shelf life is getting shorter.

![Figure 3. Changes in weight loss during cold storage](image)

3.4. Water Content

Based on figure 4, changes in water content tend to decrease during storage. Pineapple stored at cold temperatures can retain more water contained in the fruit than higher temperatures. This is because lower temperatures will inhibit the rate of respiration. The slow rate of respiration will slow down the reaction of decomposition of glucose and oxygen into carbon dioxide and water. Thus, the fruit that is stored at a lower temperature will release less water. Also, the condition of the storage room that has high humidity affected the fruit to release the water content slowly.

![Figure 4. Changes of water content during cold storage](image)

3.5. Shell and Flesh Colour

Based on figure 5, it shows that pineapple that was stored at a higher temperature will have discolouration more quickly along with storage time and maturity because high temperatures will increase respiration in the fruit so that it will cause discolouration along with maturity. The higher the temperature, the change in outer maturity will be faster. Whereas at a storage temperature of 7 °C, changes of maturity in outside fruit tend to be slow. Cold temperatures will inhibit respiration in the fruit so that changes in the physical quality of the shell colour will run more slowly. At a cold storage temperature until the end of storage, the fruit does not reach the maximum of maturity or 100% of shell colour. It means the fruit still fresh and good looking to sell in the market.
Figure 5. Changes of visual shell colour during cold storage

Figure 6. Changes of a) flesh and b) shell hue angle during cold storage

Based on figure 6, the hue angle of flesh fruit is increased, and the shell is decreased until the end of storage. It means the flesh changes from yellow colour to green and the shell from green to yellow as a process of maturity. This is due to pineapple stored at low temperatures that will inhibit the rate of respiration and maturity so that the pineapple fruit colour changes slowly. The slight increase in hue angle changes is caused by the characteristics of pineapple flesh, which qualitatively changes the colour that is getting pale due to cold storage. Pineapple stored at 7°C has a higher hue angle increase in the flesh of the fruit. It is suspected that the fruit stored at very low temperatures will cause the chilling injury, and the colour of the flesh turns to pale. Figure 6 showed the visual of whole fruit and half-cut fruit. The visual changes from 0-day storage to 20-day storage also observed black spots, and the flesh colour turns to pale identically caused by the chilling injury.

Figure 7. The visual of a) shell 0-day b) shell 20-days c) flesh 0-days and d) flesh 20-days.
3.6. Acidity
Based on figure 8 showed that changes in the acidity of pineapple during storage tends to decrease. It is suspected that cold temperatures cause chilling injury in pineapple during storage. It is because of the pineapple is very susceptible due to the cold storage. The application of refrigeration for pineapples is limited due to the development of chilling injury symptoms at temperatures below 13 °C [22].

![Figure 8. Changes of acidity during cold storage](image)

3.7. Total Dissolved Solids
The total dissolved solid increases during storage. During the ripening process, the fruit will become sweeter after organic acids are converted into sugar, which can reach the proportion of 20 % in ripe fruit. Pineapple stored at 7°C will continue to form sugar until the end of storage because it does not go through a phase of quality loss, so the TDS will continue to increase [23].

![Figure 9. Changes of TDS during cold storage](image)

3.8. Hardness
Fruits stored at cold storage result in a lower decrease in hardness compared to fruits stored at higher temperatures. This is caused by storage at higher temperatures will result in higher respiration rates and faster enzyme activity. The texture of the fruit will be softer when fruit turn to more mature. Respiration rate of fruits stored at low temperatures tends to be slow. This is because low temperatures inhibit the rate of respiration. Thus, the fruit ripens slowly, and the texture does not soften quickly.
4. Conclusions
Overall, changes in respiration and physical quality of pineapple occurred slowly during cold storage. Pineapple crownless has the lowest respiration rate at 7°C the rate is 2.05 mlO$_2$.kg$^{-1}$.hour$^{-1}$ and 5.90 mlCO$_2$.kg$^{-1}$.h$^{-1}$. The Michaelis-Menten modelling has a good fit to the observation with determination coefficient 0.85 and 0.91. The physical quality attributes change slowly during cold storage and the fruit at the end of 20-days still fresh fruit but observed chilling injury.

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5. References
[1] Mulderij R 2018 An Overview Global Pineapple Market. Accessed from www.freshplaza.com/article/2194086/overview-global-pineapple-market on 15 May 2019
[2] Tarigan H K, Yulius, M E, Nggaro, Y M, Dewi, E K and Apriyadi, T E 2014 Panduan Pascapanen Nenas. (Jakarta: Agricultural Ministry of Indonesia)
[3] Lobo M G and Paull R E 2017 Handbook of Pineapple Technology: Production, Postharvest Science, Processing and Nutrition (Chichester, UK: John Wiley & Sons)
[4] Anwar R S 2005 Dampak Kemasan dan Suhu Penyimpanan terhadap Perubahan Sifat Fisik dan Masa Simpan Brokoli Setelah Transportasi (Institut Pertanian Bogor: Bogor)
[5] Gardjito M and Saifudin, U 2011 Penanganan Pascapanen Buah-Buahan Tropis (Yogyakarta: Kanisius)
[6] Hong S and Kim D 2001 Influence of oxygen concentration and temperature on respiratory characteristics of fresh-cut green onion Int. Journal of Food Science and Tech. 36 (3), pp. 283-289
[7] Lee D S, Lee K S, Park I S and Yam K L 1994 Analysis of respiration characteristics of low CO$_2$ tolerance produces for designing modified atmosphere package Food Science and Biotechnology 3, pp. 99–103.
[8] Yam K L, Haggar P E and Lee D S 1993 Modeling respiration of low CO$_2$ tolerance produce using a closed system experiment Food Science and Biotechnology 2, pp. 22–25.
[9] Benitez S, Chiumenti M., Sepulcre F, Achaerandio I and Pujolá M 2012 Modeling the effect of storage temperature on the respiration rate and texture of fresh cut pineapple Journal of Food Engineering 113 pp.527–533.
[10] Fonseca S C, Oliveira F A R and Brecht, J K 2002 Modelling respiration rate of fresh fruits and vegetables for modified atmosphere packages: a review Journal of Food Engineering 52 (2) pp. 99–119.
[11] Mannapperuma J D and Singh R P 1994 *Modeling of gas exchange in polymeric packages of fresh fruits and vegetables* Ed. Singh R P, Oliveira F A R Process optimization and minimal processing of foods (New York: Elsevier)

[12] Cameron A, Talasila P and Joles D 1995 *Predicting film permeability needs for modified-atmosphere packaging of lightly processed fruits and vegetables* HortScience: a publication of the American Society for Horticultural Science 301 (1) pp. 25–34

[13] Bhande S D, Ravindra M R and Goswami T K 2008 *Respiration rate of banana fruit under aerobic conditions at different storage temperatures* Journal of Food Engineering 87 pp. 116–123

[14] Mahajan P V and Goswami T K. 2001 *Enzyme kinetics based modelling of respiration rate for apple* J. Agric. Eng. Res 79 pp. 399–406

[15] Hagger P E, Lee D S and Yam K L 1992 *Application of an enzyme kinetics based respiration model to closed system experiments for fresh produce* Journal of Food Process Engineering 15 (2) pp. 143–157

[16] Lee J J and Lee D S 1996 *A dynamic test for kinetic model of fresh produce respiration in modified atmosphere and its application to packaging of prepared vegetables* Journal of Food Science and Biotechnology 5 pp. 343–348

[17] Mangaraj S and Goswami T K 2011 *Measurement and modeling of respiration rate of guava (CV. baruipur) for modified atmosphere packaging* International Journal of Food Properties 14 (3) pp. 609–628

[18] Song Y, Kim H K and Yam K L 1992 *Respiration rate of blueberry in modified atmosphere at various temperatures*. Journal of the American Society for Horticultural Science 117 pp. 925–929

[19] McLaughlin C P and O’Beirne D 1999 *Respiration rate of a dry coleslaw mix as affected by storage temperature and respiratory gas concentrations* Journal of Food Science 64 (1) pp. 116-119

[20] Henig Y S and Gilbert S G 1975 *Computer analysis of the variables affecting respiration and quality of produce packaged in polymeric films* Journal of Food Science 40 pp. 1033-1035

[21] Beaudry R M, Cameron A C, Shirazi A and Dostal-Lange D L 1992 *Modified atmosphere packaging of blueberry fruit: effect of temperature on package O₂ and CO₂* Journal of the American Society for Horticultural Science 117 pp. 436-441

[22] Brown B I 1986 *Temperature management and chilling injury of tropical and subtropical fruit* Acta Horticurae 175 pp. 339-342

[23] Simon E J, Dickey J L, Reece J B and Hogan K A 1999 *Campbell Essential Biology with Physiology* (Addison: Wesley)