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Respiration rate, ethylene production and chlorophyll content of the fruit and crown of pineapple stored at low temperatures

(Kadar respirasi, pengeluaran etilena dan kandungan klorofil buah dan jambul nanas yang disimpan pada suhu rendah)

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Key words: pineapple, crown, respiration rate, ethylene production, chlorophyll content, storage, chilling injury

Abstrak

Perubahan kadar respirasi, pengeluaran etilena dan kandungan klorofil pada keseluruhan buah, bahagian buah sahaja (tanpa jambul) dan jambul nanas kv. Gandul yang matang semasa dan selepas penyimpanan pada pelbagai suhu telah dikaji. Bahagian buah (tanpa jambul) dan bahagian jambul berbeza-beza dari segi kadar respirasi. Kadar respirasi bahagian-bahagian lain buah turut dipengaruhi oleh suhu dan tempoh simpanan. Kadar pengeluaran etilena amat rendah bagi semua bahagian buah. Bahagian buah mengalami kecederaan dingin apabila disimpan melebihi 3 minggu pada suhu 5 °C. Kerosakan jambul berlaku pada semua suhu dan bukan merupakan kecederaan dingin. Kandungan klorofil jambul dan kulit berkurangan semasa penyimpanan pada suhu 10 °C dan selepas pendedahan ke suhu 28 °C (ambien). Kandungan klorofil pada jambul tidak semestinya menunjukkan tahap kesegarannya.

Abstract

The changes in the respiration rate, ethylene production and chlorophyll content in mature whole fruits, fruit body (fruit without the crown) and crown of pineapple cv. "Gandul" during and after storage at various temperatures were studied. The fruit body and the crown behaved differently to the rate of respiration. The respiration rate of different portions of the fruit was influenced by temperature and length of storage. The ethylene production was very low in all portions of the fruit. The fruit body was affected by chilling injury when stored for more than 3 weeks at 5 °C. Crown deterioration took place at all temperatures and was not a form of chilling injury. The chlorophyll content of the crown and skin decreased during storage at 10 °C and after holding at 28 °C (ambient). The chlorophyll content of the crown did not necessarily indicate its freshness.

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Introduction

Pineapple is classified as a non-climacteric fruit which exhibits continuous low respiration and ethylene production rates during ripening (Dull 1971; Kader 1992). The pineapple fruit is made up of two major parts namely the fruit body and the crown. The fruit body is a composite fruit comprising many small fruitlets attached to the core (Dull 1971). There is a maturity gradient from the bottom to the top portion of the fruit. The fruitlets at the lower level are more mature than the upper ones close to the crown (Tay 1976; Ramlah 1981). The crown comprises leaflets, usually green in colour. Since the two major parts are very different in their physical characteristics, therefore their physiological behaviour and responses to environmental changes at post harvest are also expected to be different, especially with regard to chilling injury.

Chilling injury in horticultural crops has been comprehensively reviewed by Jackman et al. (1988) and Wang (1990). In pineapple, chilling injury is characterised by the failure of the green shell to turn yellow, the yellow shelled fruit turning to a dull or brown colour, wilting, drying and discolouration of crown leaflets and a breakdown of internal tissue, resulting in a pale watery appearance (Dull 1971). Paull and Rohrbach (1985) suggested that the pineapple crown is more susceptible to chilling injury than the fruit itself. The occurrence of crown deterioration was believed to be associated with chilling injury due to exposure to temperatures below the optimum level for storage. However, no detail studies have been reported. Since there is a variation in morphological and physiological properties of different portions of pineapple fruit, Abdullah and Rohaya (1997) proposed that storage problems of fruit body and the fruit crown should be addressed and tackled separately.

There is a need to study the physiological behaviour including the rates of respiration and ethylene production of the different parts of the fruit for a better understanding of the whole system. The present study reports some of the changes in respiration and ethylene production rates of the whole fruit, fruit body and the crown of pineapple cv. Gandul stored at low temperatures. Changes on the chlorophyll content of the crown leaflets and the skin as results of low temperature storage are also discussed.

Materials and methods

Fruits

Pineapple cv. Gandul at mature green stage were obtained from Lee Pineapple Plantation, Simpang Renggam, Johor. The fruits were transported to MARDI's Postharvest Laboratory at Serdang, Selangor immediately after harvest. The good quality and uniform fruits were selected for the study. The peduncles of the fruits were trimmed with a sharp knife leaving a remaining length of approximately 2–3 cm. The peduncle ends were then dipped in 500 ppm benomyl (*Benlate* 50% a.i.) to control diseases especially black rot caused by *Thielaviopsis paradoxa* (Lim 1985).

Crown appearance, skin colour and chlorophyll content

Gandul pineapple were stored at 5, 10, 15 and 20 °C in telescopic corrugated fibreboard boxes. Each box contained six fruits. A total of 360 fruits per storage temperature were studied. The fruits were removed weekly from the low temperatures until 5 weeks storage. The skin colour and crown changes were evaluated on all fruits immediately after removal and 6 days after holding at ambient temperature (28 °C).

The crown appearance was rated from 1–5 following the scale below:

1 = fresh and green
2 = good with the tip ends very slightly yellow
3 = moderately good, tip ends slightly yellow or brown and dry

4 = not good, tip ends dry with more intense yellowing or browning
5 = yellow or brown and dry

The skin colour was rated from 1–6 as follows:
1 = mature green
2 = breaker
3 = quarter ripe
4 = half ripe
5 = three-quarter ripe
6 = fully ripe

The chlorophyll contents were evaluated only on fruits stored at 10 °C. For this purpose, two fruits from each box (total of six boxes in each observation) were selected randomly. The crowns were divided into the top, middle and bottom portions. The crown leaflets from each portion were cut into very small pieces. The fruit body was also divided into the top, middle and bottom portions. The skin from each portion was peeled at the base level of the "eyes". Twenty grammes of the sample was used for extraction and analysis. Chlorophyll from the leaflets and the skin was extracted and analysed spectrophotometrically using spectrophotometer model UV-160 1 PC Shimadzu (Ranganna 1977).

Respiration and ethylene production rates

The fruits were divided into two groups. The first group was left intact while the fruits of the second group were cut carefully into the fruit body and the crown. Besides the peduncle end, the cut areas separating the fruit body and the crown were also dipped into 500 ppm benomyl to control postharvest diseases. The whole fruit including the fruit body (without the crown) and the detached crown (without the fruit body) were then placed individually in respiratory jars and stored at 2, 5, 10, 15 and 24 °C in three replicates.

For the first week, gas measurements were conducted on every alternate day, after which measurements were carried out weekly until the fourth week. Fruits stored at 24 °C were measurable only until the third week. For each determination, the jars were closed for 2 hours.

Respiration rate was measured based on the production of carbon dioxide under static system. Gas samples were extracted from the atmosphere inside the enclosed jar with a hypodermic syringe. The gas was injected into a Varian 1420 gas chromatograph fitted with a thermal conductivity detector and a stainless steel column of 150 cm x 3 mm packed with 80–100 mesh Porapak R for carbon dioxide determination. The carrier gas for carbon dioxide determination was helium with a flow rate of 30 mL/min and a column temperature of 35 °C.

For ethylene determination, 1 mL of the respired gas was injected into a Varian 1440 gas chromatograph fitted with a flame ionization detector and a stainless steel column of 180–120 mesh Porapak T. The carrier gas for ethylene determination was nitrogen at a flow rate of 30 mL/min and a column temperature of 100 °C.

Statistical analysis

The data were statistically analysed with analysis of variance (ANOVA). Significant differences among means were detected using Duncan Multiple Range Test (DMRT).

Results and discussion

Crown freshness

The freshness of crown was well maintained for 5, 4, 2 and 1 week(s) in fruits stored at 5, 10, 15 and 20 °C respectively *(Table 1)*. After the respective periods at 10, 15 and 20 °C, the crown scores increased which indicated that crown deterioration process was taking place under the respective storage environments. The changes in crown scores did not take place at all at 5 °C. When the fruits were held further at ambient temperature, crown scores increased faster which indicated that crown deterioration process was accelerated at ambient temperature after storage at all temperatures. Crown deterioration at ambient (28 °C) was

Table 1. Effects of low temperatures on crown scores of Gandul pineapple upon removal from storage and after holding for 6 days at ambient

Time of inspection	Storage period (week)	Crown score*			
		5 °C	10 °C	15 °C	20 °C
Upon removal	0	1.0c	1.0d	1.0a	1.0c
	1	1.0c	1.0d	1.0a	1.0c
	2	1.0c	1.0d	1.0a	2.3b
	3	1.0c	1.0d	1.5b	2.3b
	4	1.0c	1.0d	1.8b	2.6b
	5	1.0c	2.3c	**	**
6 days at ambient	0	1.0c	1.0d	1.0c	1.0c
	1	1.0c	1.0d	1.7b	1.2c
	2	2.0b	3.0b	3.8a	3.4a
	3	3.0a	3.0b	**	**
	4	3.0a	5.0a	**	**
	5	**	**	**	**

Each value is the mean of six replicates. Values with the same letters in the same column are not significantly different at 5% level according to DMRT
*Crown scores:
1 = fresh and green
2 = good with the tip ends very slightly yellow
3 = moderately good, tip ends slightly yellow or brown and dry
4 = not good, tip ends dry with more intense yellowing or browning
5 = yellow or brown and dry
**Fruits had already spoilt

observed in fruits previously stored at all temperatures. The higher the temperature during storage, the faster the fruit discolouration at ambient after removal. The degree of the deterioration was not determined by the temperature where the fruits were previously stored. It can be concluded that crown deterioration is not a form of chilling injury since it took place at all temperatures. Crown deterioration is a probable natural senescent process of the fruit and not a physiological disorder. This conclusion can be drawn since chilling injury is time-temperature related (Wilkinson 1970). Chilling injury can develop after enough exposure to temperatures below the optimum level for storage.

Changes in chlorophyll contents in the pineapple crown during storage at 10 °C and after being held for another 6 days at ambient are shown in *Table 2*. The initial chlorophyll content was highest in the middle portion followed by the top and the bottom. The chlorophyll content in all crown portions dropped significantly after low temperature storage and declined further as storage was extended. Continuous decline in chlorophyll also took place at holding temperature. Chlorophyll is known to be higher in mature leaf and lower in young leaf or shoot. As the leaf become older, chlorophyll decreases due to degradation process. It is a natural phenomenon that chlorophyll in plant parts degraded after harvest and during storage. Apparently, the chlorophyll contents of the leaflets did not necessarily indicate freshness since the leaflets with fresh appearance after prolonged storage also contained low chlorophyll. For instance, the fruits stored at 5 °C remained fresh and green for up to 5 weeks storage but the chlorophyll content dropped significantly as storage period was extended.

Table 2. Chlorophyll contents in 3 sections of the crown of Gandul pineapple during storage at 10 °C and after holding for 6 days at ambient following low temperature storage

Time of inspection	Storage period (week)	Chlorophyll (mg/100 g fresh weight)		
		Top	Middle	Bottom
Upon removal	0	2.77a	3.83a	1.37a
	1	1.24b	1.31b	0.23b
	2	0.99b	1.23b	0.31b
	3	1.01b	1.23b	0.27b
	4	0.63b	0.92b	0.16b
	5	0.55b	0.74b	0.14b
6 days at ambient	0	1.13b	1.12b	0.16b
	1	1.13b	1.40b	0.36b
	2	0.69b	0.86b	0.16b
	3	0.54b	0.66b	0.15b
	4	**	**	**
	5	**	**	**

Each value is the mean of six replicates. Values with the same letters in the same column are not significantly different at 5% level according to DMRT
**Fruits had already spoilt

Skin colour changes

The fruit remained green throughout storage of 5 weeks at 5 °C *(Table 3).* Skin colour development took place slowly at 10 °C with skin yellowing first observed after 4 weeks. At 15 °C and 20 °C, skin colour developed more rapidly since yellowing had already been observed on stored fruits as early as 1 week after storage.

Colour development progressed further when the fruits previously stored at 10 °C and above were held further for 6 days at ambient. Normal ripening process at ambient also took place in fruits stored previously for less than 2 weeks at 5 °C. However, fruits stored for more than 3 weeks at 5 °C failed to ripen satisfactorily. These fruits developed a brownish colour on the skin which indicated the occurrence of chilling injury on the fruit body (Dull 1971). According to Hardenburgh et al. (1990), the optimum storage temperature for pineapple is in the region of 7–13 °C. Abdullah (1999) recommended the optimum storage temperature for Malaysian pineapples at between 8–10 °C.

The development of chilling injury of the fruit body of Gandul pineapple at 5 °C after 3 weeks is therefore unavoidable. The development of chilling injury symptoms in the fruit body, but not in the crown leaflets at 5 °C confirms that the two portions of the fruit have different tolerance levels to low temperatures. This was suggested earlier by Abdullah and Rohaya (1997). Apparently, the fruit was more chilling sensitive than the crown of Gandul. This is not in agreement with the suggestion made by Paull and Rohrbach (1985) that the crown is more sensitive to chilling than the fruit itself. The level of chilling sensitivity of the crown can also be influenced by the cultivar. Different degrees of susceptibility to chilling injury among cultivars have been reported in many fruits including bananas (Pantastico et al. 1990).

Besides yellowing of the skin, the ripening of the fruit body was also accompanied by the decline of skin chlorophyll (Dull et al. 1967). For fruits stored at 10 °C, the decline of skin chlorophyll is clearly shown in *Table 4*. The reduction in skin chlorophyll was observed

Table 3. Effects of low temperature storage on skin colour score development of Gandul pineapple

Time of inspection	Storage period (week)	Colour score*			
		5 °C	10 °C	15 °C	20 °C
Upon removal	0	1.0c	1.0d	1.0d	1.0d
	1	1.0c	1.0d	1.3d	1.8cd
	2	1.0c	1.0d	2.1c	5.4ab
	3	1.0c	1.0d	5.4a	6.0a
	4	1.0c	2.1c	5.6a	6.0a
	5	1.0c	2.1c	**	**
6 days at ambient	0	1.5bc	1.5cd	1.5cd	1.5c
	1	2.5b	3.2b	4.5b	4.7b
	2	3.1a	5.7a	4.5b	6.0a
	3	1.6c***	5.9a	**	**
	4	1.0c***	5.9a	**	**
	5	**	**	**	**

Each value is the mean of six replicates. Values with the same letters in the same column are not significantly different at 5% level according to DMRT
*Colour score:
1 = mature green
2 = breaker
3 = quarter ripe
4 = half ripe
5 = three-quarter ripe
6 = fully ripe
**Fruits had already spoilt
***Symptoms of chilling injury were observed

Table 4. Chlorophyll contents in 3 sections of the skin of Gandul pineapple during storage at 10 °C and after holding for 6 days at ambient following low temperature storage

Time of inspection	Storage period (week)	Chlorophyll (mg/100 g fresh weight)		
		Top	Middle	Bottom
Upon removal	0	0.62a	ND	ND
	1	0.61a	0.60a	0.55a
	2	0.52ab	0.44b	0.44b
	3	0.40bc	0.47b	0.44b
	4	0.33de	0.30c	0.28c
	5	0.22de	0.17d	0.17d
6 days at ambient	0	0.53a	0.44b	0.43b
	1	0.30cd	0.17d	0.10d
	2	0.05ef	0.04e	0.05d
	3	0.11f	0.07e	0.10d
	4	0.10f	0.06e	0.05d
	5	**	**	**

Each value is the mean of six replicates. Values with the same letters in the same column are not significantly different at 5% level according to DMRT
ND = Not done
**Fruits had already spoilt

both during storage at low temperatures and after holding at ambient in all portions of the fruit. The temperature of 10 °C did not cause chilling injury. Therefore, the association of chlorophyll content with chilling injury of the skin could not be concluded from this study.

Respiration rates

The respiration rates of the whole fruit, the fruit body and the crown during storage at 2, 5, 10, 15 and 24 °C are shown in *Figures 1–3* respectively. For the whole fruit *(Figure 1),* the respiration rates at 2 °C and 5 °C were maintained at very low levels between 3 and 6 mL CO_2/kg/h throughout storage. The respiration rates at these two temperatures showed a slight increase during the earlier stage of storage and declined towards the end. Similar trends were also observed for whole fruits stored at 10 °C and 15 °C but the respiration rates were in a higher range of 5–9 mL CO_2/kg/h. At 24 °C, respiration rates were maintained around 12 to 14.5 mL CO_2/kg/h during the first week but jumped to more than 25 mL CO_2/kg/h on the second week. This rate was maintained until the third week.

The respiration rate of the fruit body was also determined by the temperature *(Figure 2)*. At 2 °C and 5 °C, the respiration rates were maintained at very low levels between 4–6 mL CO_2/kg/h. At other temperatures, the respiration rates showed an increasing trend throughout storage. The respiration rates were higher at higher temperatures. However, the trends were completely different with detached crowns *(Figure 3)*. At 2, 5 and 10 °C, the respiration rates decreased during the first 3 days. After this period, the respiration rates increased until the curves reached the peak before declining to lower levels. At 15 °C, respiration rates declined sharply until the fifth day before increasing. The increase in the respiration rates took place until the second week before declining. At 24 °C, respiration rates dropped sharply during the first five days, followed by a slow decrease until the third week.

The results showed that the crown and the fruit body of pineapple have different respiration rates. The respiratory pattern of the fruit body was almost similar to the whole fruit since the weight of the fruit body represents about 85% of the whole fruit weight. Kader (1992) classified pineapple as a fruit with a low respiration rate. The classification is based on the range of respiration rate between 5–10 mg CO_2/kg/h (2.4–4.8 mL CO_2/kg/h) measured at 5 °C. The results from this study showed

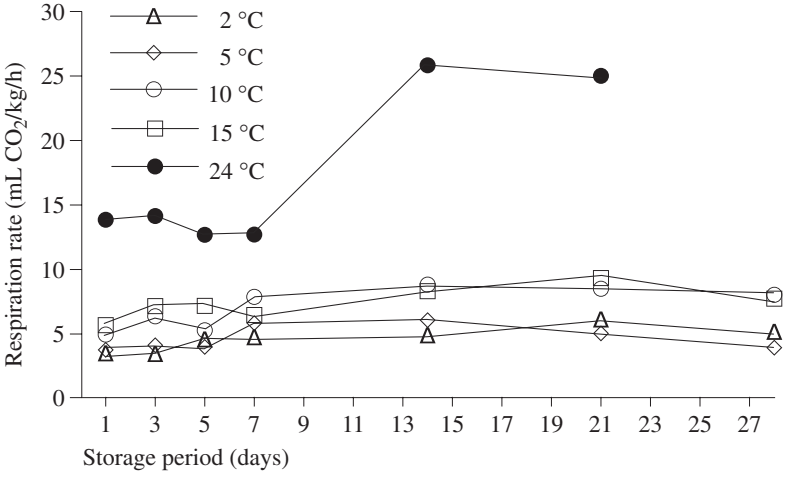

Figure 1. Respiration rates of the whole fruit of pineapple cv. Gandul during storage at different temperatures

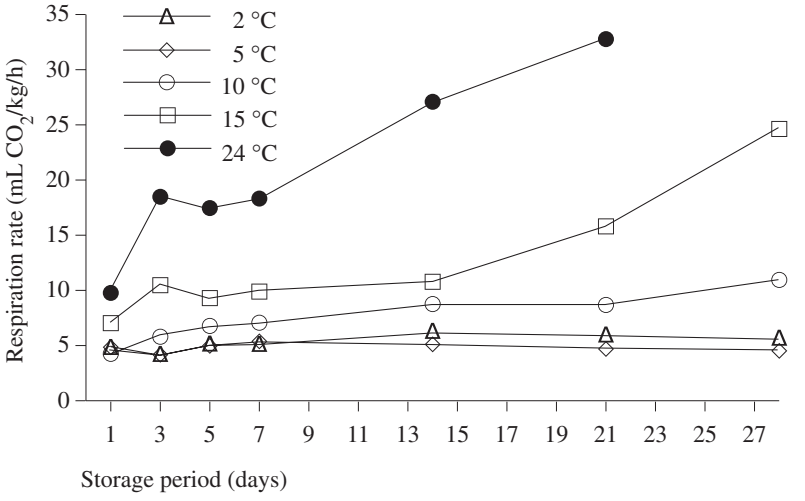

Figure 2. Respiration rates of the fruit body of pineapple cv. Gandul during storage at different temperatures

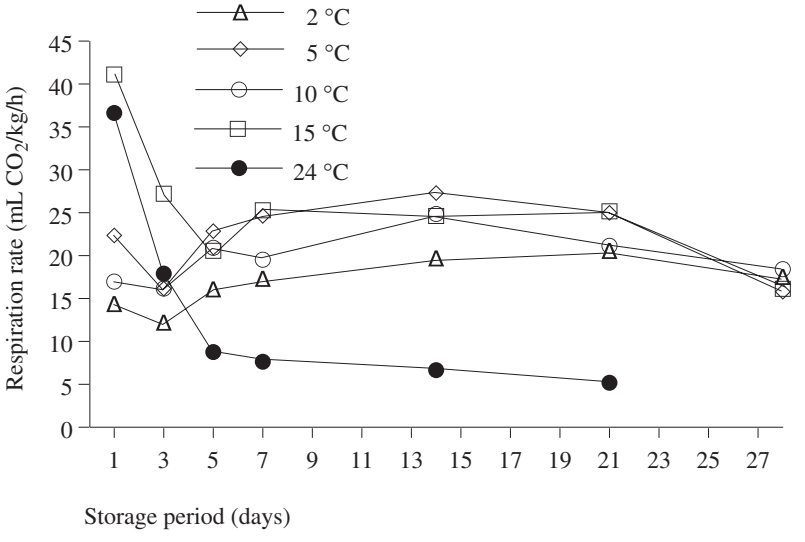

Figure 3. Respiration rates of the crown of pineapple cv. Gandul during storage at different temperatures

that this classification is only valid for the whole fruit and the fruit body but not the crown since the respiration rates of the crown were much higher. Under the classification method by Kader (1992), it is more appropriate to classify the crown in the high category together with some leafy vegetables. High respiration rates have been reported on many leafy vegetables including celery (Berg and Lenz 1972), lettuce and spinach (Scholz et al. 1963).

Ethylene production

The rates of ethylene production of the whole fruit, fruit body and the crown during storage at all temperatures were very low. The rate is close to zero most of the time for the whole fruit. Similar results were observed in the detached fruits (without the crown) at all temperatures, except at 24 °C. At 24 °C, ethylene production was traced during the first 3 days and on the third week of storage. Ethylene production of more than

2 mL/kg/h was observed in crowns after 3 days at 15 °C and 24 °C but generally the ethylene production was very low or did not take place most of the time.

The results demonstrated that the ethylene productions in the whole fruit or in different portions of the Gandul pineapple are very low. These results are in agreement with Dull et al. (1967) who reported very low production of ethylene in whole Smooth Cayenne pineapple. Other fruits with low ethylene production rates include blackberry, blueberry, cucumber, eggplant, okra, pepper, persimmon and watermelon (Kader 1992). Leafy vegetables are also included in this category (Kader 1992). This is in agreement with the low ethylene production rate in the crown of pineapple from this study.

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