Respiration, Ethylene and Color Changes during Ripening of 'TN-2' and 'Mex' Papaya (*Carica papaya* L.)

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Keywords: Respiration rate, Ethylene production, Coloring

Summary

The objective of this research was to investigate the change in color, ethylene production, and respiration of 'TN-2' and 'Mex' papaya fruit during ripening. The data showed that ethylene production and respiration rate of 'TN-2' papaya started from day1 and 'Mex' started from day 6 for ethylene production and day 9 for respiration rate. The comparison between color changes and ethylene production showed that 25% yellowed skin of 'TN-2' began to accumulate ethylene. In 'Mex' papaya at 50% yellowed skin began to accumulate ethylene.

Introduction

Carica papaya L. was a tropical fruits which origin in Central America. Fruits itself contain abundant of carotenoids, vitamin C and papain. Moreover, according to their ripening behavior, papaya was classified into climacteric fruits which can produce respiration and ethylene peak during ripening period. At the same time, fruits will soften rapidly (Chao, 2010; Lazan *et al.*, 1989). Moreover, carotenoid accumulation and increased in soluble glucose and fructose (Gomez *et al*., 2002; Zhou *et al*., 2001; Schwieggert *et al*., 2011). In spite of that, declination of phenolic compound also be found during ripening (Laura *et al*., 2010)

Ethylene was one of the plant hormone which could affect plant growth and development such as seed germination, flower and leaf senescence, biotic or abiotic stress, and fruit ripening. During activation, ethylene bound to the receptor around the endoplasmic reticulum called

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ethylene receptor (ETR) as the first-defense line and activated the downstream signal, constitutive triple response (CTR) which was a negative regulator for downstream signal, ethylene insensitive (EIN). Further activation, EIN induce ethylene response factor (ERF) which directly bound to DNA sequences (Stepanova and Ecker, 2000). According to Yang (1985) in ethylene biosynthesis, L-methionine transformed into S-adenosyl methionine (SAM) and converted into 1-aminocyclopropane-1-carboxylic acid (ACC) and ethylene by the help of three important enzymes: ATPase, ACC synthase and ACC oxidase.

Zhang (1990) observation in different genotypes of papaya showed different behavior in ethylene production and respiration rate during ripening period. In addition, these ethylene and respiration changes between cultivars affected the fruit's firmness (Manenoi, 2007). Currently, there are several cultivars had commercially planted such as: 'Maradoll', 'Golden', 'Sunrise', 'TN-1', 'TN-2', 'TN-5', 'TN-6', and 'Redlady' with different characteristic among them (Oliveira, 2011; Sanudo-Barajas, 2009; Wang, 2005; Crane, 1995). The aim of this study was to investigate the ripening action in respiration rate, ethylene and color changes between 'TN-2' and 'Mex' (Mexico breeding line) papaya cultivars.

Material and Methods

25% yellowed skin of 'Mex' (Mexico breeding line) and 'TN-2' papaya cultivars were harvested from non-commercial farm at Taichung with 3 replicate each treatments to examine their color changes, ethylene production, and respiration rate.

1. Color observation

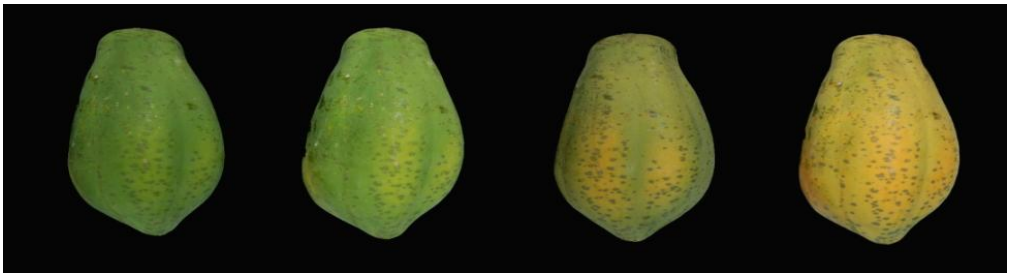
The color alteration of fruits during ripening period were marked with 1-4 where 1=0-25% yellowed skin, 2=26-50% yellowed skin, 3=51-75% yellowed skin, and 4= 76-100% yellowed skin (Fig. 1).

2. Ethylene production

Each of the papayas was put into 4L chamber with air flow 4L/liter. The observation was done everyday till fruits showed a climacteric peak. 1 ml of air was collected from every sample by syringe and injected into Gas Chromatograph, Shimadzu Model GC-8A with injection port, column and detector was at 130, 90, and 130^{0}C. The ethylene' standard was 1.34 ppm. The calculation of the ethylene production:

Ethylene (µl C_2H_4/kg hr):

[(sample peak(cm) x attenuation)-(air peak(cm) x attenuation)]x std conc (ppm) x flow rate(L/hr) /(std peak (cm) x weight (kg))



0-25% 25-50% 50-75% 75-100%

1 2 3 4

Color index

Fig. 1. Papaya color index

3. Respiration rate

The examination of respiration rate used flow system method where sample was put into 4L chamber with air flow 4L/hr. The carbon dioxide detection was done every day until climacteric peak was shown. Each sample collected 1 ml of air by syringe and injected into IR-analyzer, Maihak, Model UNOR610. Standard of carbon dioxide was 1.486%. The calculation:

Respiration rate (CO_2/kg hr):[(sample peak (cm) – air peak (cm)) x std conc (%) x 10 x flow rate (L/hr)] / (std peak (cm) x weight (kg))

Results

1. Color changes

Comparison of color changes between 'Mex' and 'TN-2' papaya cultivars during ripening period were investigated. In 'TN-2' papaya maintained its color at 25% yellowed skin about 1 day then began to change rapidly at next days and reached fully ripen at the forth day. In contrast, 'Mex' papaya fruit maintained at 25% yellowed skin for 2 days then increased slightly till 50% at the third and fourth day. Last, 'Mex' papaya fruits began to increase drastically at the fourth day and reached fully ripe at eightieth day (Fig. 2).

2. Ethylene production

In 'TN-2' papaya, ethylene production started increasing from first day and till the end of the investigation. Compared with 'Mex' papaya, ethylene production was below 0.01µl C2H4/kg

hr till day 5. The ethylene production began to increase at day 6 and reached the highest peak at day 9 and maintained at same fluctuation till the end of the observation (Fig. 3).

3. Respiration rate

To compare the respiration rate between 'TN-2' and 'Mex' papaya fruit during ripening. The results showed that the respiration of 'TN-2' papaya was lower than 'Mex' papaya about 5.98 and 9.72 CO2/kg hr where respiration rate of 'TN-2' increased till the end of investigation. In contrast, 'Mex' papaya's respiration rate maintained in range at 8.09 to 11.09 CO2/kg hr till day 9 and started to incline to highest peak about 17.88 CO2/kg at day 10 and then declined for next two days (Fig. 4)

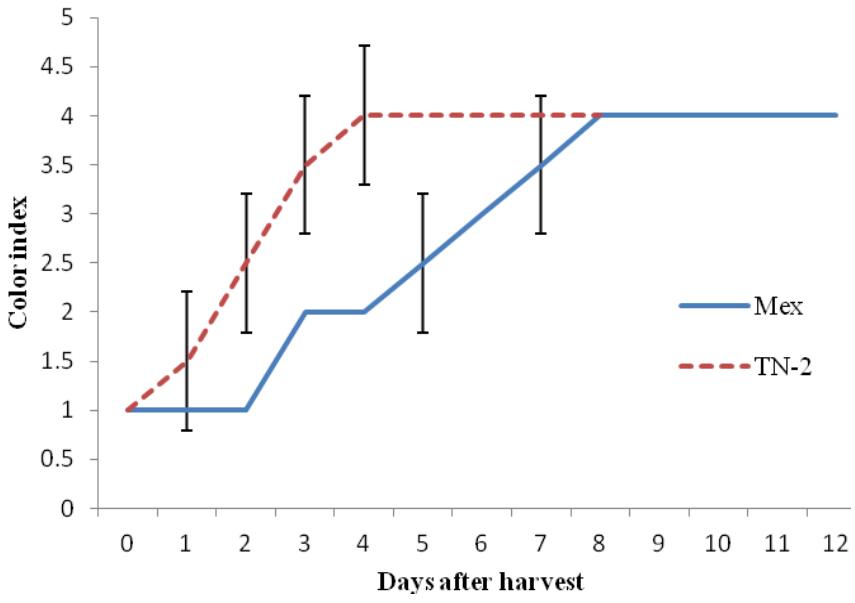


Fig. 2. Comparison of color changes between 'Mex' and 'TN-2' papaya during ripening.

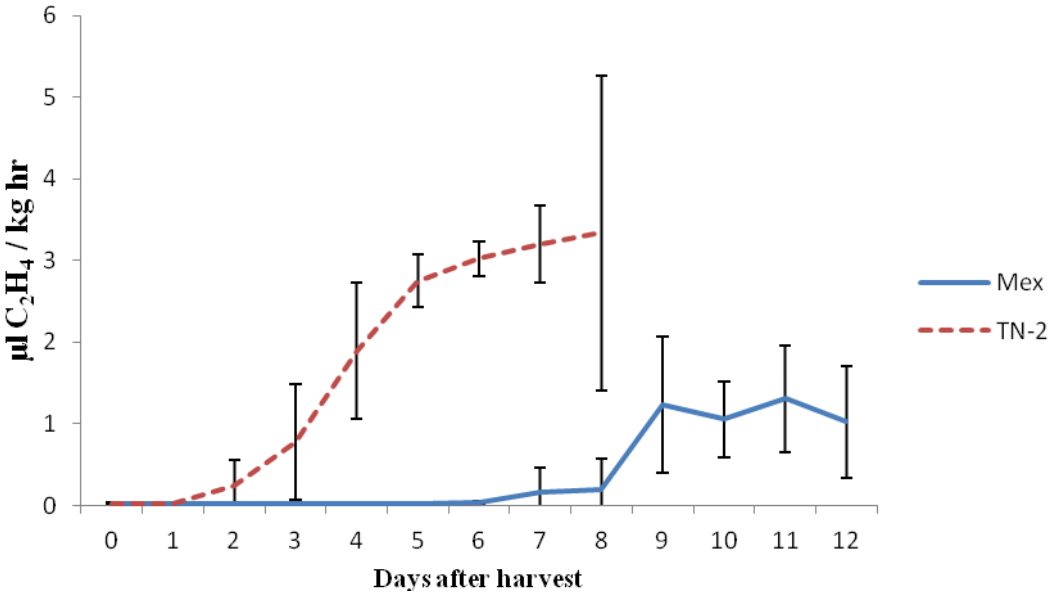


Fig. 3. Comparison of ethylene accumulation between 'Mex' and 'TN-2' papaya during ripening.

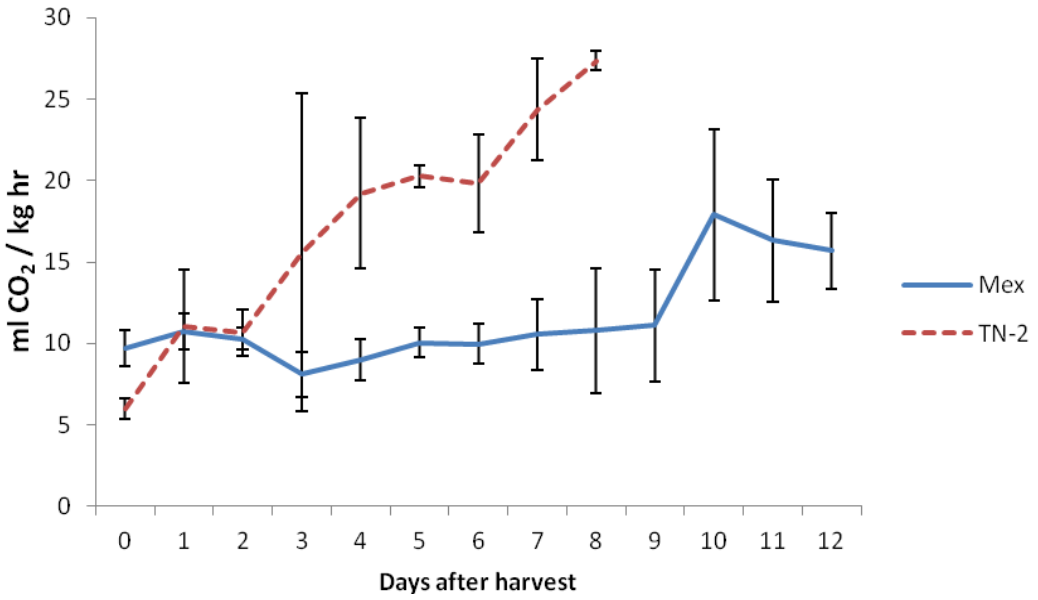


Fig. 4. Comparison of respiration rate between 'Mex' and 'TN-2' papaya during ripening.

Disscussion

Accoding to their response to ethylene, fruits could devided into two types: climacteric and non-climacteric fruits where their diferences were at ethylene preception which control the physiological ripening. Yang (1987) research, climacteric fruit ethylene action could divide into two types where immature fruits lack of system II and mature fruits had both receptors: system I and system II. Comparison between color changes and ethylene production, the results (Fig. 2 & 3) showed that 'TN-2' papaya's climacteric peak started at 25% yellowed skin and 'Mex' papaya started from 50% yellowed skin.

The inclination of respiration rate also could be found in both cultivars (Fig.: 4) which suggested there was close correlation between ethylene biosynthesis since oxygen was needed in conversion of ACC to ethylene and carbon dioxide by the helped of ACC oxidase (Yang, 1985). Moreover, Plant senescence was shown by the decreasing of plant's ability to respond in stress or failed to do its metabolism like the inactivation of cytochrome oxidase resulting the accumulation of electron in complex III (cytochrome reductase) in mitochondria (Dufour *et al*., 2000). The leak of electron to oxygen could result the production of reactive oxygen species (ROS). Wang (2010) observation, in salt stress showed that ethylene could induce plant alternative oxidase pathway and suggested that alternative oxidase might play an important role at inhibition of ROS production (Maxwell *et al*., 1999).

Color changes could also find during ripening period and had close correlation with chlorophyll degradation. At the early step of chlorophyll metabolism, chlorophyll breakdown could through two ways: Chlorophyllase (CHL) way which dephytylated the chlorophyll into chlorophyllide and phytol then removal of Mg^{2+} by Mg-dechelatase into pheophorbide; Pheophytinase (PPH) way which first chlorophyll's Mg^{2+} was removed by Mg-dechelatase become pheophytin then dephytylation of pheophytin into pheophorbide a (Pheide a) and phtyol (Schelbert *et al*., 2009 and Tanaka and Tanaka, 2006). Next the Pheide a converted into red chlorophyll catabolite (RCC) and fluorescent chlorophyll catabolite (FCC) by pheophorbide a oxygenase (PAO) and red chlorophyll catabolite reductase (RCCR) (Tanaka and Tanaka, 2006). The conversion of the Pheide a into RCC is the last check point of green lost (green to red) (Barry, 2009). Shimolawa (1978) research in citrus showed that the ethylene could induce the chlorophyllase activity.

Schelbert (2009) showed that PPH was the major factor in dephytylation during senescence. This result was concomitant with Zhang (2011) and Buchert (2011) during investigation of leaf senenscence in Chinese flowering cabbage ((*Brassica Rapa Var.* Parachinensis) and broccoli by using cytokinin and ethylene found that an induction of chlorophyllase activity and gene

expression. However, the hue angle and the chlorophyll content in cytokinin and ethylene treatment experiment showed that cytokinin treatment maintained higher green color and chlorophyll content than control and ethylene treatment (Buchert *et al*., 2011). In PPH, gene expression increased which following by the decreasing chlorophyll content and color changes in ethylene treatment (Buchert *et al*., 2011). Other chlorophyll degradation related enzymes like Mg-dechelatase, PAO and RCCR also found to be induced by ethylene (Zhang *et al*. 2011). This phenomenon suggested that chlorophllase function was limited regulation via posttranslational regulation (Harpaz-Saad *et al*., 2007). Other possibility that affected the color changes was the stay-green gene induction which involved in light harvesting complex II disassembly in thylakoid membrane (Hortensteiner, 2009). During color changes investigation (Fig.: 2) showed different color changes in both papayas suggested that stay-green gene had been induced and PPH might be induced by other signaling pathway except ethylene (Buchert *et al*., 2011). For further degradation, PAO and RCCR also play an important role in chlorophyll metabolism. The deficiency of the PAO and RCCR could result the cell death activation (Lorrain, 2003) which was triggered by accumulation of Pheide a and RCC (Hortensteiner and Krautler, 2011) and supposed to be related with cell death signaling or defense pathways (Lorrain, 2003).

In brief, 'TN-2' papaya began to form climacteric peak started from 25% yellowed skin and climacteric peak 'Mex' papaya started from 50% yellowed skin.

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'台農二號'和'Mex'番木瓜(*Carica papaya* L.)果實在後熟期間的呼吸率、乙烯生合成及果皮轉色之變化

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摘要：本試驗探討'台農二號'和'Mex'番木瓜在後熟期間果皮轉色、乙烯釋放率和呼吸率變化之關係。結果顯示，'台農二號'番木瓜的乙烯釋放率和呼吸率在第一天即開始上升，而'Mex'番木瓜的乙烯釋放率和呼吸率分別在第六天和第九天才開始上升。番木瓜'台農二號'在 25%轉黃階段時，果皮轉色指標隨著乙烯生合成速率上升而增加；然而，番木瓜'Mex'則是在 50%轉黃階段時，果皮轉色指標才隨著乙烯生合成速率上升而增加。

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