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Colour at harvest and post-harvest behaviour influence paprika and chilli spice quality.

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Abstract

Capsicum annuum L. paprika and cayenne chilli pepper fruit were grown for red spice production and harvested at various colour stages on the same day. Fruit of each stage were allowed to change colour at room temperature with or without the addition of 100 μl l^{-1} ethylene. Fruit appearance and colour development, and respiration and ethylene production were measured during the colouring period. Ethylene treatment had no effect on colour development or pungency for both cultivars, even though it easily crossed the cuticle, epidermis and flesh tissues into the fruit cavity. Green or deep green harvested fruit failed to fully colour red, while fruit that were harvested at or after the colour break stage visually completed their red colour development within 7–9 days. However, the colour intensity of spice powder was low for all fruit that had not developed a deep red colour prior to harvest. For paprika no difference between deep red fruit that were succulent or that had partially dried on the plant was found, but chilli fruit that had partially dried before harvest produced the most intense colour. American Spice Trade Association (ASTA) extractable red colour was the best measure of spice colour quality, compared to reflected lightness (L*), chroma (C*) and hue angle (h°) colour measurements. Pungency did not change between ripeness stages for chilli and was absent in paprika. Paprika and chilli fruit showed climacteric behaviour as long as they were attached to the plant, but when detached were non-climacteric. © 2000 Elsevier Science B.V. All rights reserved.

Keywords: *Capsicum annuum* L.; Ethylene; Respiration; Colour; Pungency; Spice

1. Introduction

Worldwide interest in capsicum spices is increasing (Bosland, 1993). These are either pungent or non-pungent spices, such as pimiento, paprika, and chilli that are produced from dried fruit and are ground into powders. To achieve a good spice colour quality, completely red fruit are processed.

Since fruit in different positions on the chilli or paprika plant mature at different times in the growing season, cost-effective once-over mechanical harvesting yields a mixture of fruit of different ripeness stages. Pre- or post-harvest treatments may increase the number of red ripe fruit by

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inducing fruit ripening (Saltveit, 1977; Gomez, et al., 1998). However, results from these treatments are often unsatisfactory (Knavel and Kemp, 1973; Krajayklang et al., 1999). While preharvest studies indicate ethylene involvement in colour development (Osterli et al., 1975), overall information on the ripening behaviour of harvested *Capsicum* species is limited.

Fruit in the genus of *Capsicum* have been classified as non-climacteric (Lurie et al., 1986; Biles et al., 1993), but the hot chilli cv. 'Choorae-hong' (*Capsicum frutescens*) was reported as climacteric (Gross et al., 1986). By definition, in non-climacteric fruit an increase in respiration and ethylene production during ripening is absent (Biale, 1964). Treatment with exogenous ethylene in climacteric fruit such as banana leads to an autocatalytic biosynthesis of ethylene and this accelerates fruit ripening, but ethylene synthesis does not occur spontaneously in non-climacteric fruit after treatment with exogenous ethylene (Biale, 1948).

The aim of this study was to investigate the effects of colour stage at harvest and ethylene on paprika and chilli fruit colour changes to improve colour quality, pungency, and yield of red fruit after once-over harvesting.

2. Materials and methods

2.1. Plant material

Healthy fruit of PS72285 paprika (*Capsicum annuum* L.) and Caysan SPS705 chilli (*Capsicum annuum* L.) were hand-harvested on the same day at different colour stages. Seven colour stages were harvested; light green, deep green, breaker (slight colouration), breaker red (some red colour), bright red (100% red), deep red and succulent, and deep red and partially dried. Ten fruit of each colour category were randomly selected and separated into two groups, one as a control and one for ethylene treatment. The experiment was replicated three or four times for paprika or chilli, respectively.

2.2. Ethylene treatment and storage conditions

Within one hour after harvest, five fruit were weighed in bulk and enclosed in 2.2 l plastic containers with 5 g of calcium hydroxide ($Ca(OH)_2$) to absorb evolved CO_2. Ethylene was injected through a septum port into the sealed ethylene C_2H_4 treatment containers, to obtain 100 $\mu l\ l^{-1}$. Control containers were vented every 12 h for 5 min, followed by ethylene containers to prevent cross-contamination. Ethylene was then re-injected until 48 h. Thereafter, containers were opened and fruit stored in the same container in a well-ventilated room at 22°C under normal fluorescent room light for 7–10 days depending on their external appearance. Visual colour, weight loss, CO_2 and C_2H_4 evolution were determined during storage.

2.3. External and internal quality assessments

Skin colour was assessed daily on a subjective scale from 0 (light green) to 11 (deep red and partially dried), using a modified scale from Lownds et al. (1994). External quality was judged at the end of storage based on fruit skin shrivelling (water loss) and calyx yellowing. Decay incidence was also evaluated at the end of storage as percent of fruit manifesting stem-end rots or other rots, and on the inside of cut fruit.

2.4. Colour and pungency determination of spice

After quality evaluation and gas measurements, fruit were dried to constant weight in a hot air oven at 45°C and ground with a Culatti electric mill (1.5×10^{-8} m mesh size). Ground samples were kept in airtight plastic bags in the dark at room temperature and used for colour and pungency measurements.

2.4.1. Reflected colour

Surface colour of sample powder was measured as reflected colour in the CIELAB (L*a*b*) colour space using a Minolta model CR-300 Colorimeter (Minolta, Osaka). One reading was performed for each sample. Lightness L*, chroma C* and hue angle h° were determined, with L* ranging from 0 = black to 100 = white. C*, $((a^*)^2 + (b^*)^2)^{1/2}$) and represents colour saturation which varies from dull (low value) to vivid colour (high value), and h° = $\tan^{-1}$ (b*/a*) and is defined as a colour wheel, with red-purple at an angle of 0°, yellow at 90°, bluish-green at 180°, and blue at 270° (McGuire, 1992).

2.4.2. Extractable colour

Extractable red colour was measured in the units of the American Spice Trade Association (ASTA) (Woodbury, 1997). A representative ground sample (~ 70–100 mg) was extracted in 100 ml of acetone for 16 h at room temperature in the dark. Absorption of this solution was measured at 460 nm in comparison to a standard glass reference. The final ASTA value for each measurement was calculated on a dry weight basis as previously described by Krajayklang et al. (1999).

2.4.3. Pungency

The pungency of each sample was estimated by determining the gross capsaicin and dihydrocapsaicin content (Todd et al., 1977), using a high performance liquid chromatographic (HPLC) procedure measuring absorbance at 280 nm (Krajayklang et al., 1999). Scoville values were calculated by multiplying the amount of capsaicin and dihydrocapsaicin per gram dry weight by 16 million Scoville heat units for pure capsaicin and dihydrocapsaicin (Todd et al., 1977).

2.5. Gas measurements

Rates of C_2H_4 and CO_2 production were measured daily by gas chromatograph. A static, closed system was employed, whereby the storage container for the above fruit was sealed for a set period at 22°C before taking gas samples for assessment.

2.5.1. C_2H_4 measurement

To measure C_2H_4, a 1-ml gas sample was collected from the container after 4 h of sealing the containers. C_2H_4 was quantified using a Varian chromatograph model 3400 equipped with a flame ionisation detector (Varian Australia, Mulgrave, Vic.) and a Porapak Q stainless steel column (60 cm × 3.1 mm i.d.) of 80/100 mesh. Temperature conditions were 50°C for the column, 135°C for the injector and 150°C for the detector. Flow rates of the carrier gas nitrogen, air and hydrogen were 50, 300 and 40ml/min, respectively. A gas standard containing 95 nmoles l^{-1} C_2H_4 standard (BOC Gases, Torrensville, SA) was used for calibration. Results were expressed as nmoles of ethylene produced per kg fresh weight and h (nmoles.kg^{-1} h^{-1}).

2.5.2. CO_2 measurement

CO_2 was measured at hourly intervals for 4 h by injecting another 1-ml gas into a Varian 3300 chromatograph equipped with a thermal conductivity detector (Varian Australia, Mulgrave, Vic.) and a silica column (35 cm × 3.1 nm i.d.) of 80/100 mesh. Temperature conditions were 28°C for the column and 90°C for the injector and detector, and the flow rate of the carrier gas helium was 5ml/min. A calibration was performed using a 0.5% CO_2 standard (BOC Gases). Results were expressed as mmoles of CO_2 produced per kg of fresh weight and h (mmoles.kg^{-1} h^{-1}).

2.5.3. Ethylene uptake by fruit

The internal C_2H_4 concentration in the fruit cavity was measured in contrast to the external one surrounding the fruit, in order to determine fruit ethylene uptake. Two groups of five fruit were enclosed in separate containers. One was used for a control (no C_2H_4 injection) and the other for a 100 μl l^{-1} C_2H_4 treatment. C_2H_4 was injected into the sealed container, and it was left at room temperature for 12 h. Two 1-ml gas samples were taken from a container, and used to determine the external C_2H_4 concentration. Internal fruit atmosphere samples were taken by inserting the needle of a 1 ml syringe through the ovary wall into the fruit cavity. C_2H_4 concentrations were determined as above.

2.6. Statistical analyses

The experiment was conducted as a completely randomised factorial design, with seven colours at harvest × two ethylene applications. Experiments were replicated using 3–4 different harvests over a month period. Each cultivar was evaluated as a separate experiment. Statistical analyses were performed using Genstat 5 for Windows Release 4.1 (3rd edition, Rothamsted Experimental Station, England). Data were subjected to two ways analysis of variance, and treatment means were compared using least significant differences ($P < 0.05$).

3. Results

3.1. Fruit appearance

External quality of the green and the deep green harvested fruit after storage was very poor for both primarily from significant shrivelling, but that of the breaker to the red harvested fruit was acceptable after 10 days of storage. Ethylene had no effect on quality or fungal spoilage in paprika or chilli (data not shown).

Harvested chilli fruit of all colour stages developed a slight discolouration on the seed after storage, indicating possible internal fungal contamination, while harvested paprika fruit showed no sign of any fungal development throughout the experiment. There was no visible disease for any fruit after storage.

3.2. Fruit colour development

Ethylene application did not influence the final colour of fresh paprika and chilli fruit (Table 1). However, maximum colour for paprika was slightly delayed by ethylene application.

Green and deep green harvested fruit of both cultivars achieved less than 50% red colouration during storage (Table 1), even after ethylene treatment. Up to 8 days additional exposure to ethylene did not further promote colour development of these fruit (data not shown). Fruit harvested at

Table 1
Colour development of *Capsicum* fruit harvested at different colour stages during storage at room temperature with or without ethylene application

Treatment factor		Final colour[z]	Time to final colour (days)
Paprika cv. PS72285			
Colour at harvest	Green	4.4b[y]	8.2abc
	Deep green	4.7b	9.3a
	Breaker	10.2a	8.7ab
	Breaker red	10.2a	7.7bc
	Bright red	10.4a	7.0c
	Deep red	11.0a	3.5d
	Deep red + dried	11.0a	0.0e
Ethylene	Without (−)	8.9a	5.9b
	With (+)	8.8a	6.7a
Chilli cv. Caysan SPS705			
Colour at harvest	Green	2.1e	9.5a
	Deep green	4.3d	9.0a
	Breaker	9.4c	8.9a
	Breaker red	9.8bc	8.8a
	Bright red	10.8ab	8.5a
	Deep red	11.0a	4.9b
	Deep red + dried	11.0a	0.0c
Ethylene	Without (−)	8.3a	6.8a
	With (+)	8.4a	7.3a

[z] Skin colour was scored on a scale from 0 to 11: 0, green; 1, deep green; 3, 25% red; 5, 50% red; 7, 75% red; 9, bright (full) red; 10, deep red and succulent; 11, deep red and partially dried.

[y] Different letters within columns for each treatment factor and cultivar show significant differences ($P < 0.05$) using LSD.

Table 2
Reflected and extracted colour, and pungency of *Capsicum* powder made from fruit that were harvested at different colour stages and ripened with or without ethylene

Treatment factor		Colour characteristics[z]				Pungency ($\times 10^3$ SHU)[y]
		L*	C*	h°	ASTA	
Paprika cv. PS72285						
Colour at harvest	Green	52a[x]	52b	69a	50d	0
	Deep green	52a	57b	65b	63d	0
	Breaker	45b	66a	58c	104c	0
	Breaker red	43b	70a	54d	139b	0
	Bright red	42bc	71a	54cd	139b	0
	Deep red	41bc	68a	53d	169a	0
	Deep red + dried	38c	67a	51d	194a	0
Ethylene	Without (−)	45a	64a	58a	123a	0
	With (+)	45a	65a	57a	123a	0
Chilli cv. Caysan SPS705						
Colour at harvest	Green	52a	44c	79a	25f	16a
	Deep green	52a	53b	70b	38e	16a
	Breaker	47b	71a	56c	74d	17a
	Breaker red	46b	71a	56c	89c	17a
	Bright red	45b	72a	55c	90c	18a
	Deep red	42c	71a	54c	106b	17a
	Deep red + dried	42c	70a	53c	120a	22a
Ethylene	Without (−)	47a	64a	60a	76b	18a
	With (+)	46a	65a	60a	79a	18a

[z] Lightness (L*) ranged from 0 = black to 100 = white, chroma (C*) = $(a^2+b^2)^{1/2}$ with 0 = least intense, hue angle (h°) = $\tan^{-1}(b/a)$ with 0° = red-purple and 90° = yellow in the CIELAB colour space. American Spice Trade Association (ASTA) colour units measured extractable red colour/g dry weight.

[y] Scoville heat units (SHU) were calculated by multiplying the amount of capsaicin and dihydrocapsaicin per gram dry weight by 16 million Scoville units for pure compounds.

[x] Different letters within columns for each treatment factor and cultivar show significant differences ($P < 0.05$) using LSD.

breaker stage or later developed a dark red colouration (stage 10), except for breaker chilli fruit that only turned bright red (stage 9). Fruit harvested at breaker to bright red stage reached their final colour within seven to nine days (Table 1).

3.3. Colour of spice powder

Reflected colour of the powder was not affected by ethylene treatment for either cultivar (Table 2).

As colour stage at harvest increased, paprika powder of redder fruit obtained a darker colour as indicated by a reduction of L* value, from 52 to 38, for the green and deep red and partially dry harvested fruit, respectively (Table 2). Colour saturation (C*) increased for green to breaker-harvested fruit, and then remained constant (Table 2). The higher C* value for red harvested fruit represents a more vivid colour (Table 2). The hue angle of green harvested fruit after storage describes a more yellow colour with the highest angle of 69, while an increase in colour stage at harvest resulted in an increase in red colour with the lowest angle of 51 in red harvested fruit (Table 2).

Powder colour of chilli fruit showed the same characteristics. With increasing colour stage at harvest, powder of stored fruit was more dark, vivid and deep red in colour. The L* values ranged from 52 to 42, C* ranged from 44 to 70, and hue angle was about 70–53 for green harvested fruit to deep red harvested fruit, respectively (Table 2).

Deep-red coloured fruit at harvest (stage 10 or 11) achieved the maximum extractable red colour, 194 and 120 ASTA units for partially dry paprika and chilli fruit, respectively (Table 2). There was no difference in extractable colour intensity between powders made from succulent and partially dried paprika fruit, but for chilli, letting fruit partially dry on the bush increased extractable colour intensity of its powder. Green and deep green fruit at harvest obtained a very low colour intensity of 50–60 ASTA units for paprika and about 25 to 40 ASTA units for chilli (Table 2). Breaker to bright red fruit at harvest, while appearing visually as red or deep red after storage, had reduced extractable red colour of the spice powder.

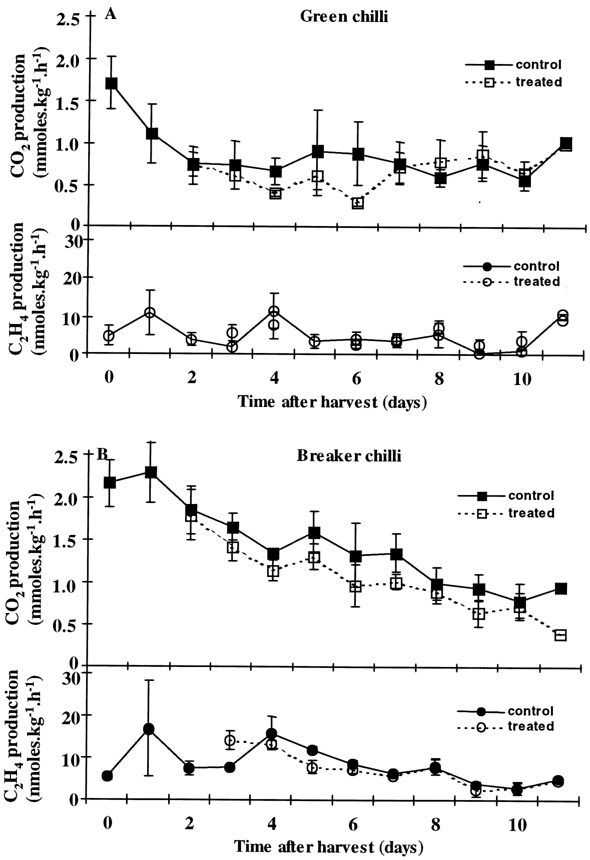

Fig. 1. Ethylene and carbon dioxide production at 22°C of green mature (A) and breaker (B) harvested chilli fruit exposed for 48 h to air (control) or 100 μl l^{-1} ethylene (treated). Four replications of five fruit were used for each measurement; error bars represent the SE of the mean.

3.4. Pungency

Pungency or hotness was absent in paprika fruit. Pungency of chilli powder did not increase significantly with colour stage at harvest (Table 2). Ethylene had no effect on pungency (Table 2).

3.5. CO_2 and C_2H_4 production

3.5.1. During post-harvest storage

Typical data for respiration and ethylene production of chilli fruit are shown in Fig. 1, as the behaviour of chilli and paprika fruit was very similar. In general, neither respiration nor ethylene production of green or breaker harvested fruit was affected by exogenous ethylene treatment, but these data were not available during ethylene treatments (Fig. 1). Respiration declined directly after harvest in all fruit without a significant respiratory peak, and ethylene production did not change markedly throughout the experiment (Fig. 1). Green fruit did not reach a fully red colour before measurements concluded, whereas breaker fruit did.

3.5.2. At harvest

Typical data for respiration and ethylene production rates of different colour stages of chilli fruit immediately after harvest are shown in Fig. 2, as paprika and chilli showed very similar behaviour. A distinct respiratory climacteric pattern was apparent, with respiration peaking at dark green to breaker red colour stages (Fig. 2). Ethylene production peaked later, increasing from breaker red and peaking at the bright red colour stage (Fig. 2).

3.5.3. Ethylene uptake by fruit

At harvest, both control and treated fruit had a similar internal ethylene atmosphere concentration at 0.01–0.02 μl l^{-1}. After ethylene was injected into the container, internal and external ethylene atmosphere was markedly different between treated and untreated fruit. Ethylene levels of 0.02–0.06 and 18.2–22.8 μl l^{-1} were detected in the internal atmosphere inside the cavity of control and treated fruit, respectively. The external

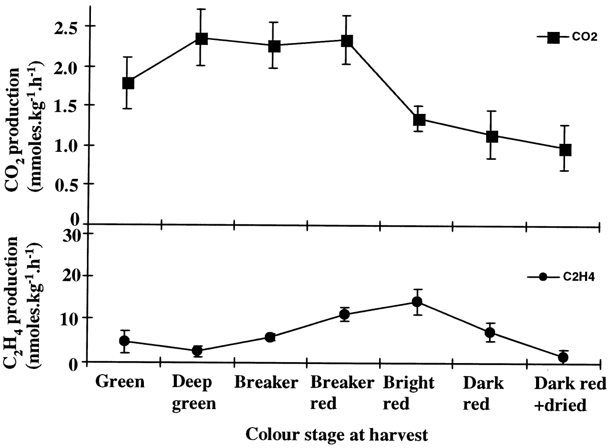

Fig. 2. Ethylene and carbon dioxide production at 22°C of differently coloured cv. Caysan SPS705 chilli fruit directly after harvest. Four replications of five fruit were used for each measurement; error bars represent the SE of the mean.

atmosphere surrounding control fruit contained 0.04–0.05 μl l^{-1} ethylene, while that of treated fruit was about double that in the fruit cavity at about 40.2–43.9 μl l^{-1}. The difference may be caused by air leakage into the syringe during sampling and after retracting the needle due to reduced pressure within the syringe and the small internal fruit volume during sampling.

4. Discussion

4.1. Fruit appearance

Green harvested fruit appeared to be more susceptible to water loss compared to red harvested fruit after 10 days of storage. It has been noted that during the growing season green chilli fruit are highly sensitive to water loss and disorders influenced by environmental conditions such as heat damage (Wall and Biles, 1994). Therefore, if fruit are to be stored for the fresh market water loss control, especially for green fruit, is essential.

4.2. Fruit colour development

Exogenous ethylene treatment was not effective in inducing red colour development of paprika and chilli fruit under the conditions of this study. Green mature harvested fruit failed to fully colour even when treated with ethylene. Similar results were found in detached green pimiento (Knavel and Kemp, 1973) or bell pepper fruit (Lockwood and Vines, 1972) after they were treated with either ethylene (500 μl l^{-1}) or ethephon (1000 μl l^{-1}). Lockwood and Vines (1972) suggested that the thick cuticle of capsicum fruit is a barrier to ethylene thus preventing its action. However, we observed that a high level of internal ethylene uptake occurred under ethylene treatment, indicating that ethylene was able to cross the cuticle. Therefore ethylene is not able to promote postharvest colour development. In our study ethylene even slightly delayed paprika fruit colour development, similarly to a previous report (Lockwood and Vines, 1972) on fruit colour development of pimiento fruit. The reason for this is unknown.

Nevertheless, once fruit were harvested at or near the breaker colour stage, they coloured normally in both cultivars. While on the plant, fruit were able to change colour satisfactory and also exhibited ethylene and respiratory peaks. Colour change involves both the degradation of chlorophyll and the *de novo* production of keto-carotenoids capsanthin and capsorubin; also xanthophylls and carotenoids are present and these often become esterified (Minguez-Mosquera and Hornero-Mendez, 1994). Therefore, red colouring on the plant appears to involve some additional factor, such as other hormones or presence of sufficient pigment precursors, that interacts with ethylene to induce full colour changes from green to deep red. In addition the chlorophyll degradation process is impaired in harvested fruit.

4.3. Colour of spice powder

The concentration of extractable colour pigment in dried paprika and chilli powder was not affected by ethylene application, mirroring the lack of effect on visual fruit colour. Extractable colour on a dry weight basis was highest in fruit allowed to dry on the bush (pre-harvest dehydration). Similar findings were reported in red harvested paprika fruit (Kanner et al., 1977) and field-dried cayenne chilli (Lease and Lease, 1956). In addition, we found similar high levels for paprika fruit that were succulent but deep red. These colour stages have been previously mentioned as optimum for harvesting spice paprika in Hungary (Markus et al., 1999) and for spice paprika and Cayenne chilli in the USA (Lease and Lease, 1956). In addition to better initial colour intensity, letting fruit partially dry improves colour retention in spice (Lease and Lease, 1956), as β-carotene is converted to more stable red coloured xanthophylls (Markus *et al*., 1999).

L*, C* and h° values indicated general changes in colour for different colour stages at harvest. However, C* and h° in general only showed differences between green or partially to fully red fruit. While L* was better able to differentiate between colour stages at harvest, these measurements did not always mirror changes in ASTA colour values. Therefore, L* values cannot be used to replace the industry standard of measuring extractable red colour using the ASTA method.

4.4. Pungency

Pungency of chilli powder in this study did not vary with different colour stages. Some reports agree with this, for example Somos (1984), but Balbaa et al. (1968) and Mathew et al. (cited in Cotter, 1980) found increases in pungency as the fruit reaches the red colour stage. As only red fruit are processed into spice, always the maximum possible pungency will be achieved for the spice as influenced by colour stage.

4.5. Respiration and ethylene production

We found no response of chilli or paprika fruit to applied external ethylene. It has been reported that an application of ethylene or propylene to climacteric fruit can stimulate both respiration and autocatalytic ethylene production, while in non-climacteric fruit exogenous ethylene stimulates respiration only (McGlasson, 1978). While our paprika and chilli fruit behaved in a non-climacteric fashion after harvest, we were not able to determine respiratory stimulation by ethylene as a CO_2 scrubber had to be included during ethylene exposure to prevent CO_2 interfering with ethylene effects. However, just after treatment application these fruit had similar rates to control fruit. Contrary to us and other reports (Lurie et al., 1986; Biles et al., 1993), Gross et al. (1986) found 'Chooraehong' chilli (*C. frutescens*) to be climacteric as they found a respiratory climacteric; however, they found no C_2H_4 peak.

When comparing the respiration and ethylene production rates of different fruit colour stages on the plant, a climacteric pattern was apparent for both chilli and paprika cultivars. Respiration peaks occurred during the initial colour change, while C_2H_4 peaks occurred later as fruit completed their colour change to red. This concurs with peaks found by Wall and Biles (1994) for just harvested fruit. It therefore appears that the fruit from our study behaved differently on and off the plant, and at this stage the reason is unclear as discussed above.

The highest C_2H_4 production level for paprika and chilli fruit was about 17 nmoles.kg^{-1} h^{-1} at 22°C in this study. This was similar to other studies (Wall and Biles, 1994), but the climacteric 'Chooraehong' chillies peaked at about 30 nmoles.kg^{-1} h^{-1} (Gross et al., 1986).

Fruit harvested at an active growth stage such as green fruit tend to have high respiration rates and it is also temporarily elevated at harvest due to the harvest wound (Kays, 1991). This was also observed in this study. Maximum CO_2 production in paprika fruit at 22°C was 3.5 mmoles.kg^{-1} h^{-1} and in chilli fruit 2.3 mmoles.kg^{-1} h^{-1}; this is comparable to other studies such as of Biles et al. (1993) and Gross et al. (1986).

5. Conclusion

Different colour stages at harvest significantly affected chilli and paprika fruit colour development and spice colour quality, but not pungency levels of chilli spice. Green or deep green harvested fruit failed to fully colour red after harvest, while fruit that were harvested at or after the breaker stage completed their colour change to fully red. Exogenous ethylene application did not affect red colour development or pungency of any colour stage at harvest. Allowing fruit of both cultivars to fully ripen and/or partially dry on the bush resulted in the maximum colour intensity after processing. Extractable red colour measured using the ASTA technique was the most suitable method of spice colour assessment, compared to reflected L*, C* and h° colour measurements. A distinct climacteric pattern was found during colour change as long as fruit were attached to the mother plant. However, once fruit were harvested, green and breaker harvested fruit behaved in a non-climacteric manner.

Therefore completely red fruit at harvest, either succulent or preferably partially dry, are needed to produce the best quality spice. After once-over machine harvesting, fruit that have not achieved this colour stage should be culled before processing.

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