



HAL
open science

Culture conditions and detachment of the fruit influence the effect of ethylene on the climacteric respiration of melon.

Jenny Bower, Paul Holford, Alain Latché, Jean-Claude Pech

► **To cite this version:**

Jenny Bower, Paul Holford, Alain Latché, Jean-Claude Pech. Culture conditions and detachment of the fruit influence the effect of ethylene on the climacteric respiration of melon.. *Postharvest Biology and Technology*, 2002, 2 (2), pp.135-146. 10.1016/S0925-5214(02)00007-8 . hal-03483608

HAL Id: hal-03483608

<https://hal.science/hal-03483608>

Submitted on 16 Dec 2021

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Culture conditions and detachment of the fruit influence the effect of ethylene on the climacteric respiration of melon

Jenny Bower ^a, Paul Holford ^{a,*}, Alain Latché ^b, Jean-Claude Pech ^b

^a Centre for Horticulture and Plant Sciences, University of Western Sydney, Hawkesbury Campus, Locked Bag 1797, South Penrith DC, NSW 1797, Australia

^b UMR 990 INP-ENSAT/INRA, Ecole Nationale Supérieure Agronomique, BP 107 Auzeville, 31326 Castanet Tolosan cedex, France

Abstract

The respiratory rise associated with ripening has been considered a defining feature of climacteric fruit. However, recent work has suggested that the respiration rate remains constant in melon fruit ripening whilst attached to the plant. To clarify the relationship between attachment and the respiratory climacteric, ethylene production and O₂ consumption were monitored during the ripening of melon fruits grown in France and Australia. All fruit allowed to ripen after being detached from the vine produced both respiratory and ethylene climacterics. However, although ethylene production increased in attached fruit, the concurrent rise in respiration was either eliminated or reduced in magnitude. A positive relationship was found between ethylene levels and respiration rate. However, for a given rate of ethylene production the stimulation of respiration was less in attached than detached fruit. The application of exogenous ethylene to melons antisensed for ACC oxidase stimulated O₂ consumption only if they were detached from the vine. These data also demonstrate that attachment to the plant inhibits the effects of ethylene on respiration. In addition, it was noticed that transgenic plants were better able to tolerate infection by powdery mildew than wild-type plants. As ethylene production was greater in attached fruit, it is further suggested that feedback inhibition of ethylene synthesis is reduced while fruit remain attached to the vine. It is concluded that the presence or absence of a respiratory climacteric in fruit attached to the vine is affected by environmental factors influencing plant development. These effects may be related to changes in gas permeance, turgor, or the supply of 'plant factors' translocated through the phloem. The results support the hypothesis that the respiratory climacteric is not an essential part of ripening, but an artefact caused by stress or detachment.

Keywords: *Cucumis melo*; Respiration rate; Ethylene; Climacteric; Plant factor; Mildew tolerance

1. Introduction

For more than 70 years, the presence or absence of a climacteric increase in respiration during ripening has been used to classify fruit (Abeles

* Corresponding author. Tel.: +61-45-701-229; fax: +61-45-701-314

E-mail address: p.holford@uws.edu.au (P. Holford).

et al., 1992; Laties, 1995). This rise in respiration, along with a corresponding autocatalytic production of ethylene, is considered a defining feature of ripening in fruits such as bananas, avocados, tomatoes and melons. At about the time of the peak in respiration, fruit become edible-ripe, with changes occurring in flesh texture, pigment and aroma production (Biale and Young, 1981). It has, therefore, been assumed that the respiratory climacteric occurs to provide energy for ripening. However, it has been known for some time that certain features of the ripening process can be separated from the climacteric. In particular, the timing of colour development relative to the climacteric peak has been altered in cantaloupe melons (Pratt and Goeschl, 1969) and 'Ailsa Craig' tomatoes (Dostal and Leopold, 1967), while softening occurred before the induction of the climacteric in 'Anjou' pears (Wang et al., 1972). These results suggest that the climacteric increase in respiration is not essential for normal ripening.

The respiratory climacteric may be an artefact of harvest. Studies with Charantais melons (Shelley and Saltveit, 1993), tomatoes (Saltveit, 1993), saskatoons (Rogiers and Knowles, 1999) and purple passionfruit (Shiomi et al., 1996) have found that the ethylene climacteric was not accompanied by a corresponding rise in respiration when the fruit remained attached to the plant during ripening. However, other work has found that ethylene and respiratory climacterics both occur as Charantais melons (Hadfield et al., 1995) and tomato fruit (Andrews, 1995) ripen on the plant, even though this process may be delayed relative to harvested fruit of the same physiological age.

There are several possible explanations for the discrepancies in the above results. Knee (1995) suggested that studies of respiration that did not find a respiratory climacteric in attached fruit might have been affected by fruit photosynthesis. Also, most studies of attached fruits have measured changes in their internal atmosphere, these being expected to closely follow rates of gas exchange (Burg and Burg, 1962). However, both ripening and harvesting of fruit are likely to alter gas permeability with consequent effects on the internal atmosphere. Furthermore, the climacteric may be influenced by a plant's environment with

factors such as water stress or nutrition affecting the size and duration of increases in ethylene production and respiration rates.

To answer some of these questions, this study examined changes in respiration during ripening in melon fruit grown under different conditions. As an aid to this process, genetically engineered melons were used that expressed an antisense gene for ACC oxidase (AS) (Ayub et al., 1996). As AS catalyses the last step of ethylene biosynthesis, ethylene production is effectively blocked in these fruit. However, the fruit remain ethylene sensitive, and will ripen when treated with exogenous ethylene (Ayub et al., 1996; Guis et al., 1997). In addition, rather than monitoring the internal atmospheres of the melons, rates of O₂ consumption by the fruit were measured using respirometers (Bower et al., 1998). Respirometers are portable, automated systems that do not require connection to a gas supply or gas chromatograph and which use containers that can be of any size or shape. They are, therefore, suitable to use in a glasshouse. These devices were used to follow the rates of O₂ demand during ripening of Charantais melons grown in France and Australia.

2. Methods

2.1. Plant material

Charantais melon plants (*Cucumis melo* cv. Reticulatus F1 Alpha) containing an antisense gene for AS (Ayub et al., 1996) as well as non-transformed, normally ripening, wild-type (WT) fruit were used. One crop was grown in Toulouse, France and a further three crops were grown at Richmond, Australia. In France, the melon plants were grown in a glasshouse during the summer of 1997 using a hydroponic system, with fruits reaching maturity in September (Trial 1). The spacing of the plants was 900 mm between the rows and 600 mm within the row: WT and AS plants were alternated. The plants were supplied with a nutrient solution at a flow rate of 4 l h⁻¹ for 1 h twice each day. The composition of the solution was based on Bernadac et al. (1996) and had a conductivity of 2.5 mS with the pH being adjusted to

5.8 with nitric acid. Cooling within the glasshouse was achieved by ventilation when temperatures rose above 30 °C: convection heaters were used to maintain temperatures above 15 °C at night and 25 °C during the day. Plants were vertically trained, female flowers were hand-pollinated and three or four fruits were kept on each individual plant. Powdery mildew was controlled by dusting with sulphur.

In Australia, the plants were grown in a glasshouse with environmental controls giving average temperatures of 25 °C during the day and 18 °C at night. Once respiration measurements began, the glasshouse was maintained at a constant temperature range of 18–23 °C. The first Australian crop (Trial 2) was planted during December 1998, with a further crop started six weeks later (Trial 3). The final crop was planted in January 2000 (Trial 4). The melons were grown in 20 l pots of standard potting mix and watered by drip irrigation (Trials 2 and 3) or capillary feed (Trial 4). An initial application of slow release fertiliser (Osmocote mini[®]) was supplemented by weekly applications of a double-strength hydroponic solution (416 ppm N, 124 ppm P, 664 ppm K) during and after flowering.

In both France and Australia, the plants were grown vertically using a trellis. This, both maximized air movement around the plants and prevented injury from abrasion. Powdery mildew control was accomplished using sulphur in Trial 1, while the more humid Australian climate necessitated weekly applications of the fungicides, Benlate[®] and Mancozeb[®]. Female flowers were hand-pollinated and date tagged at anthesis. Where necessary, fruits were thinned to no more than two per plant. At the completion of the experiments, the melons were weighed and their volume measured by immersion in water.

2.2. Gas measurements

Respiration was measured in fruit ripened both on and off the plant using respirometers. The fruits were placed in sealable containers equipped with O₂ and temperature sensors connected to a computer, as described in Bower et al. (1998). In order to allow respiration measurements while melons

were attached to the vine, a 5 mm wide slot was cut in each respirometer lid through which the stem could exit. The slit was sealed around the stem using flexible, oil-based, modelling clay. Black cloth was placed over each respirometer in order to prevent photosynthesis by the ripening melon.

Outputs from the sensors inside each respirometer were monitored continuously and recorded to file at six-minute intervals. Respiration was allowed to deplete the O₂ concentration inside each container to 19 kPa. The respirometer was then flushed with air via a one-way valve until the O₂ level rose to 20 kPa, thereby maintaining the O₂ concentration between these set points.

Ethylene was measured once the melons reached 32 days after pollination (DAP). During Trial 1, ethylene was measured twice daily by withdrawing 1 ml samples from the respirometer bowls and injecting them into a gas chromatograph. For Trials 2 and 3, the respirometers were ventilated for 5–10 min then resealed for 1 h before 1 ml samples were withdrawn. These samples were injected into either a 10S Plus digital gas chromatograph (Photovac Inc., New York, USA) or a chromatograph (Gow Mac Instrument Co., USA) equipped with a flame ionisation detector.

2.3. Experimental design

In Trial 1, six AS and six WT melon fruits were used for respiration and ethylene measurements. Once ethylene production began to rise, half of the WT melons were detached, the other fruits remained attached to the vine. Half the AS melons were also detached approximately 40 DAP. At this stage, the suture lines had smoothed out and the cheeks of the fruit had filled. Melons that were detached from the plants were placed in respirometers that remained next to the vines for the duration of the experiment.

Eight additional AS melon fruit were detached and placed in respirometers in a controlled room environment at 20 ± 2 °C. Half of the pumps used to replenish the vessel atmospheres were placed inside tanks continually flushed with air containing approximately 10 ppm ethylene. The respirometers connected to these pumps were replenished with this atmosphere for the duration of the experiment.

The exhaust from these vessels was piped away from the room to avoid exposing the control fruit to ethylene.

In Trials 2 and 3, only WT fruit were studied. In Trial 2, respiration and ethylene production were measured in three fruits attached to the vine and five harvested fruits; in Trial 3, three attached and four harvested fruits were used.

For Trial 4, six AS fruits were used. Once fully mature (40–45 DAP), three melons were detached and the O₂ consumption of all six fruits measured for three days. After this period, all fruit were treated with ethylene for a further three days. To do this, the vessels were thoroughly ventilated once each day by removing the lids for several minutes. They were then resealed and ethylene injected into each vessel to raise the internal ethylene concentration to approximately 200 ppm. At the end of the trial period, the length of the main stem of each plant was measured. The plants were then oven-dried at 80 °C for 48 h and their dry weights determined. Also, the number of fruit produced per plant during the experiment was recorded. At the end of the trial period, the length of the main stem of each plant was measured. The plants were then cut at the base, oven-dried at 80 °C for 48 h and their dry weights determined. Also, the number of fruits produced per plant during the experiment was recorded.

2.4. Calculation of oxygen demand

The rate of O₂ demand by individual melons during ripening was calculated as follows:

$$\text{O}_2 \text{ demand} = \frac{\Delta\text{O}_2 \times t \times v}{P \times w}$$

where ΔO_2 is the change in O₂ concentration (kPa) between readings, t is the number of readings per hour, v is the container volume less the melon volume (ml), P is the atmospheric pressure (kPa) and w is the melon weight (kg).

As pollination dates varied, melon fruit reached their peak levels of ethylene production on different days. Therefore, data from each of Trials 1–3 using WT melons were shifted in time so that maximum ethylene production occurred on day 4 of the ethylene climacteric.

Diurnal fluctuations in temperature during Trial 1 made it necessary to normalise the respiration data to a constant temperature. To do this, mean respiration rates at temperatures between 18 and 28 °C were calculated at 0.5 °C intervals from six replications of both WT and AS melons. Regression equations modelling the increment in respiration per degree rise in temperature were calculated using data from the afternoons and evenings, as O₂ consumption was more stable when the temperature was falling. The relationship between respiration and temperature was approximately linear over the temperature interval. This enabled respiration rates to be recalculated to give the equivalent value at a temperature of 20 °C.

2.5. Statistics

Ethylene production and respiration rates from the first three trials were compared by two-way or three-way analyses of variance, as appropriate. This was done using STATISTICA (Kernel Release 5.5A, StatSoft Inc.), with time as a repeated measure. Regression analyses comparing ethylene production with respiration rates were also performed using STATISTICA, with regression lines compared using the method of Mead et al. (1993).

3. Results and discussion

3.1. Ethylene measurements of attached and detached fruit during ripening

During Trial 1, ethylene concentrations reached up to 28 $\mu\text{l l}^{-1}$ inside respirometers containing ripening attached WT fruit (Fig. 1). Despite variability in the data due to periodic flushing of the vessels, ethylene concentrations were significantly higher inside these respirometers than in those containing detached WT fruit ($P = 0.05$). The AS melons produced negligible amounts of ethylene during the experimental period regardless of whether they were attached or detached from the vine. Therefore, only data for detached AS melons are shown (Fig. 1).

Ethylene production rates during the climacteric were also significantly higher for attached fruits in

Trials 2 and 3 ($P=0.05$) (Fig. 2). This difference was particularly obvious in melons from Trial 3, where mean rates of ethylene production rose to $1.3 \mu\text{mol kg}^{-1} \text{h}^{-1}$ in attached fruit, compared to $0.4 \mu\text{mol kg}^{-1} \text{h}^{-1}$ in detached fruit.

These results are consistent with those of Hadfield et al. (1995) who found that Charan-

tais melons ripening while attached to the plant contained higher internal ethylene concentrations than harvested fruit. A similar situation has been described for attached and detached 'Red Delicious' apples (Sfakiotakis and Dilley, 1973). Rates of ethylene production were also higher in attached saskatoons compared to harvested fruit (Rogiers and Knowles, 1999).

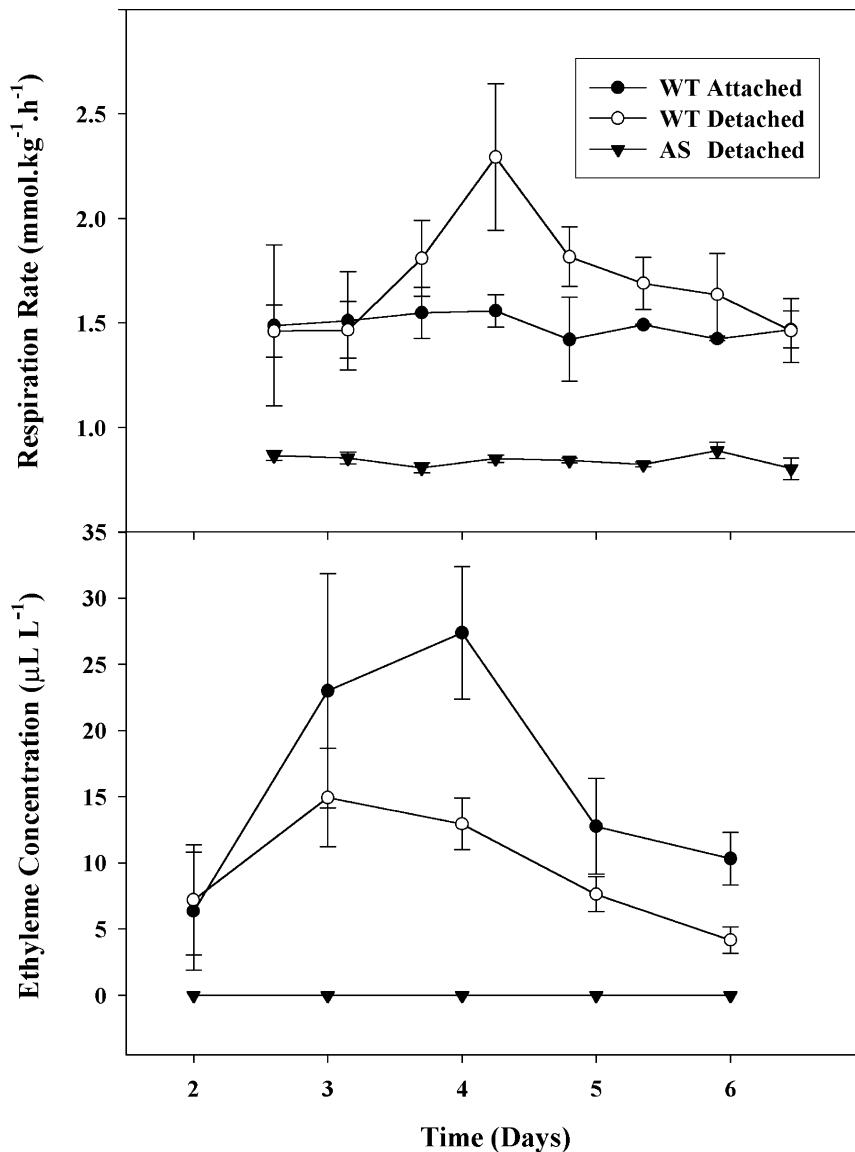


Fig. 1. Respiration rates and ethylene production during the ripening of wild-type (WT) or antisense (AS) Charantais melon either attached to the vine or removed from the plant. The plants were grown at Toulouse, France. Each data point represents the mean of three fruits. Error bars represent the standard errors of the means.

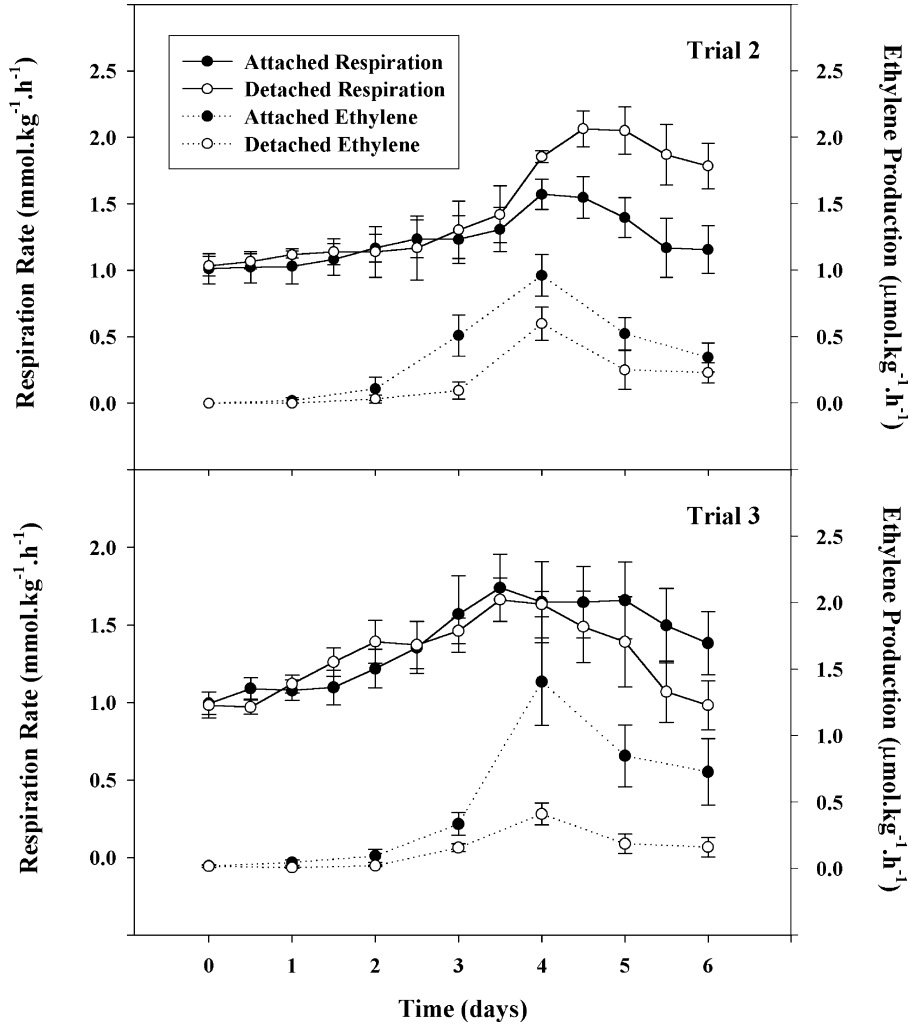


Fig. 2. Respiration rates and ethylene production during the ripening of wild-type (WT) or antisense (AS) Charantais melons either attached to the vine or removed from the plant. The plants were grown at Richmond, Australia. Each data point represents the mean of three fruits. Error bars represent the standard errors of the means.

However, other studies have not found increased ethylene production by attached compared to detached fruit. Detachment from the plant did not affect ethylene production by non-netted muskmelons (Lacan and Baccou, 1996), internal ethylene in netted melons (Shellie and Saltveit, 1993) or internal ethylene in ripening tomatoes (Saltveit, 1993). In contrast with the results of Hadfield et al. (1995), Ayub et al. (1996) found that Charantais melons

had higher internal ethylene concentrations when detached during ripening compared to attached fruit. This is consistent with the results for harvested passionfruit, which produced ethylene at a rate up to 20 times higher than fruit that ripened on the vine (Shiomi et al., 1996). Such differences are difficult to satisfactorily explain, and indicate the need for more detailed studies with consistent methods of ethylene measurement.

3.2. Rates of O₂ consumption by attached and detached fruit during ripening

It was expected that pre-climacteric rates of O₂ consumption would be similar in WT and AS melons. However, during Trial 1 it was found that respiration rates of AS fruit were significantly lower than those of WT fruit, being approximately 0.8 compared to 1.5 mmol O₂ kg⁻¹ h⁻¹ ($P = 0.05$) (Fig. 1). Detachment from the vine did not significantly affect respiration rates of the AS melons ($P = 0.05$), so data from detached AS melons are presented.

During Trial 1, detached WT melons showed the expected climacteric increase in respiration rate during ripening. This occurred concurrently with the increase in ethylene concentration inside each respirometer. In contrast, respiration rates of attached WT melons underwent little change during the experimental period despite the presence of high concentrations of ethylene (Fig. 1). Changes in colour and aroma during ripening were similar in attached and detached fruit. However, respiration rates of attached melons were significantly lower over the same period ($P = 0.05$).

All of the melons in Trials 2 and 3 underwent a respiratory climacteric during ripening, irrespective of attachment or detachment from the vine (Fig. 2). Attached melons from both planting dates had similar peak respiration rates (0.7–1.0

mmol O₂ kg⁻¹ h⁻¹). Detachment from the vine resulted in significantly faster respiration during the climacteric for melons from Trial 2 ($P = 0.05$), although this difference was not found for Trial 3 fruit. Analysis of variance of the data from these two trials showed little overall effect of trial date or attachment on respiration (Table 1). However, there was a highly significant interaction between trial, attachment and time ($P = 0.01$).

These results suggest that the growing environment may influence the development of the respiratory climacteric. The plants grown in France during the summer were produced under ideal conditions. These vines produced large, healthy melons weighing 1.2–1.6 kg each. Vines grown in Australia during the autumn were less healthy, being affected by shortened day-lengths, reduced light intensity and disease pressure. This was particularly noticeable for plants from Trial 3, which fruited during cool, wet weather in April. The melons matured more slowly than those grown in France and the ethylene climacteric commenced approximately 40 DAP rather than 35 DAP as found in Trial 1. Although the melons still developed normal colour, flavour (> 10% SSC) and aroma, they were less than half the weight of the French-grown fruit. Under these more marginal conditions, differences in respiratory behaviour between harvested and unharvested melons diminished. The health of the melon plant may therefore have had an effect on the ripening physiology of the fruit.

Table 1

Statistical significance of the main effects and their interactions from two- or three-way analyses of variance of ethylene production and respiration rates from the ripening trial conducted in France and the two trials conducted in Australia

Source	P-level			
	French data		Australian data	
	Ethylene	Respiration rate	Ethylene	Respiration rate
Trial	–	–	0.6266	0.4093
Attachment	0.0055	0.0360	0.0175	0.8167
Time	0.0208	0.0018	<0.0001	<0.0001
Trial by attachment	–	–	0.3157	0.0612
Trial by time	–	–	0.8861	0.0602
Attachment by time	0.7413	0.0114	0.0020	0.7706
Trial by attachment by time	–	–	0.1149	0.0167

Time of measurement was used as a repeated measure.

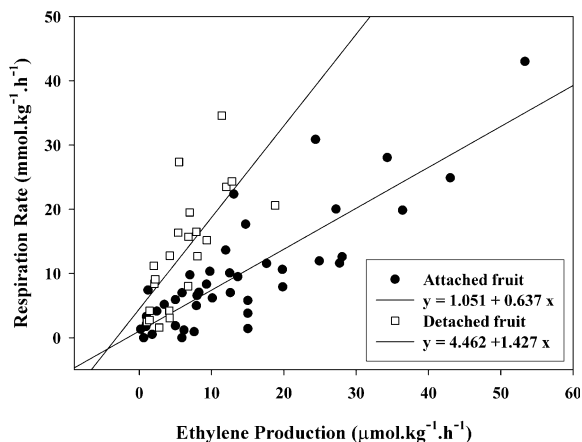


Fig. 3. Relationships between ethylene production and respiration rates from Charantais melons ripened on or off the vine. The data are combined from Trials 2 and 3 conducted in Australia.

3.3. Interactions between ethylene production and O_2 consumption by attached and detached WT fruit

Increases in ethylene production have been found to correlate closely with increased concentrations in fruit flesh (Lyons et al., 1962). In this study, the fruit allowed to ripen on the vine produced more ethylene than harvested fruit. It may be assumed that they had higher internal concentrations of the gas, as changes in internal ethylene concentration are strongly associated with increases in respiration during the climacteric (Burg and Burg, 1965). Therefore, unless the ethylene receptors become saturated, increases in internal ethylene concentration should lead to increases in respiration. Consequently, fruits attached to the vine should have had higher respiration rates and a more distinct climacteric than harvested fruits. However, the opposite was found, suggesting that attachment to the plant reduced the effects of ethylene on respiration.

To clarify the relationship between ethylene and respiration rate, increases in O_2 consumption during the climacteric were compared to ethylene production rates (Fig. 3). Comparison of the residual sums of squares from the regression ANOVA showed that the relationship between

these factors did not vary significantly between Trials 2 and 3 for either attached ($P = 0.845$) or detached ($P = 0.429$) melons and the data were combined. Further regression analysis demonstrated a significant, positive, linear relationship between the rate of ethylene production (x) and the change in respiration relative to pre-climacteric rates (y). For attached melons this could be described by the equation $y = 1.051 + 0.637 \times x$ ($r^2 = 0.70$, $P < 0.0001$), while the equation for detached melons was $y = 4.461 + 1.427 \times x$ ($r^2 = 0.51$, $P < 0.0002$). Comparison of the regression lines showed that the relationship between ethylene production and respiration rate was distinctly different for attached and detached melons ($P < 0.001$). Therefore, melons that remained attached to the vine during ripening had a smaller increase in respiration for each unit increase in ethylene production relative to detached melons.

Although the results from Trials 2 and 3 initially appeared to be different to those from Trial 1, the trends in all trials were similar. That is, attachment to the vine reduced the effects of ethylene on respiration. This was despite higher levels of ethylene production by fruit ripening while attached to the plant. Also, it appears that as the plants become more stressed from adverse cultivation conditions, then the increase in respiration for each unit of ethylene produced by the fruit is reduced.

3.4. Effects of ethylene on O_2 consumption rates of AS melons

Applying ethylene to detached AS melons during Trial 1 induced similar changes in colour and aroma as had been observed for ripening WT fruit. O_2 consumption was significantly increased by exposure to $10 \mu\text{l l}^{-1}$ ethylene ($P = 0.05$) (Fig. 4). This confirmed the sensitivity of AS melons to exogenous ethylene.

The results from Trial 4 showed that detachment from the vine did not significantly affect the rate of O_2 consumption by AS melons ($P = 0.01$) (Fig. 4). Treating the fruit with exogenous ethylene significantly increased the respiration rate of detached melons. However, O_2 consumption by the attached fruit remained unchanged ($P = 0.01$).

In this case, the stimulation of respiration by ethylene was almost completely prevented by attachment to the vine. These results support the hypothesis that attachment to the plant inhibits the effects of ethylene on respiration rates.

3.5. Vine growth of AS and WT melon plants

There was a distinct difference between the WT and AS melon plants grown in Trial 4 in relation to their tolerance of powdery mildew infection and its effect on the growth of the plant. The main stems of the AS melon plants were significantly longer than those of the WT vines throughout their growth (2.61 ± 0.16 m vs. 1.59 ± 0.10 m; $P = 0.01$). Also, each of the 15 AS vines produced at least one fruit over the experimental period, whereas the 10 WT vines produced only two fruits in total, both of which weighed less than 200 g. The AS vines had significantly greater dry weight (46.6 ± 3.6 vs. 30.4 ± 2.4 g; $P = 0.05$) than the WT vines, being $> 50\%$ heavier on average.

The reduced susceptibility of AS melon plants to powdery mildew was an unexpected result. Differences between AS and WT vines were not apparent in France, where disease pressures were less. Plant tissues normally produce ethylene rapidly in response to pathogen attack (McGlasson, 1970). Most often, ethylene is considered to have a tonic effect on plant tissues and increases

their ability to combat pathogens (Boller, 1990). However, Elad (1990) found that ethylene or methionine applied to infected plant tissues accelerated disease development after inoculation with *Botrytis cinerea*, and that inhibitors of ethylene action had the opposite effect. Mullins et al. (2000) also found that inactivation of ethylene receptor sites significantly reduced disease symptoms on grapefruit infected with *Penicillium digitatum*. Further experimentation looking at the interactions between ethylene, plant defense mechanisms and physiology is needed to clarify the role of this hormone in pathogenesis.

3.6. Potential causes of alterations in fruit physiology due to detachment

Charantais melon fruit had consistently lower rates of ethylene production when they were detached from the vine during ripening. This may have been due to limited availability of compounds required for ethylene synthesis. In attached fruit, such compounds would have been translocated to the fruit throughout ripening. An alternative explanation is that increased respiration in response to ethylene inhibits further ethylene production. By reducing ethylene sensitivity, feedback inhibition of ethylene production is also reduced. Romani (1987) suggested that increased respiration in response to ethylene is a homeostatic response to stress. It would appear from these results that attachment of melon fruit to the vine reduces ethylene related stress. Abdi et al. (1998) showed that plum fruit treated with 1-MCP, a compound that blocks ethylene perception, blocked the ability of pre-climacteric fruit to respond to applied propylene. Therefore, it appears that the respiration rate in fruit is modulated by its ability to produce and perceive ethylene.

Attachment to the plant could be analogous with the effects of the compound 1-MCP, which has been used to reduce the ethylene sensitivity and extend storage life of various commodities (Jiang et al., 1999). One side effect of MCP use has been to significantly increase ethylene production in fruit at some developmental stages (Golding et al., 1998; Mullins et al., 2000). This is

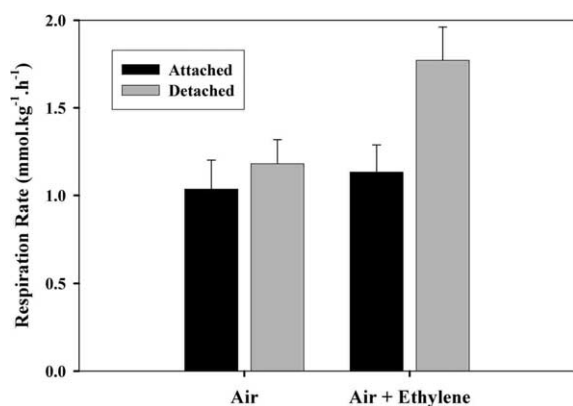


Fig. 4. The effects of ethylene treatment and attachment to, or detachment from, the vine on the respiration rate of melon fruit produced by plants containing an antisense gene for ACC oxidase.

thought to occur because MCP blocks the ethylene receptors, thus preventing normal feedback inhibition of continued ethylene synthesis (Mullins et al., 2000). If attachment to the plant also blocks some of the effects of ethylene, this could likewise affect ethylene synthesis.

Detachment from the plant has been shown to accelerate ripening in apples (Smock, 1972; Sfakiotakis and Dilley, 1973; Yang et al., 1986; Pooter et al., 1989; Blanpied, 1993), passionfruit (Shiomi et al., 1996), plums (Abdi et al., 1997) and some melon varieties (Pratt et al., 1977; Bliss and Pratt, 1979; Hadfield et al., 1995). 'Hass' avocados provide an extreme example of this phenomenon, as they are unable to ripen while attached to the tree (Adato and Gazit, 1974). The reasons for this effect are still not understood, although clearly there are a number of physiological differences between attached and detached fruit. In detached fruit, turgor is likely to fall, gas permeance may increase and the translocation of biochemical factors from the plant is interrupted. Any or all of these factors may affect the ripening process.

Turgor is unlikely to have significantly influenced the development of the climacteric in this case. Melons were kept inside respirometers, in an atmosphere that would have approached 100% RH. Detachment of a fruit from the plant involves wounding. The creation of this wound may significantly increase gas permeance of some fruit, particularly those with otherwise relatively impermeable cuticles. For example, most gas permeation has been shown to occur through the calyx scar in tomatoes (Cameron and Yang, 1982), capsicums (Burg and Burg, 1965; Bower et al., 2000) and avocados (Ben-Yehoshua et al., 1963). Burg and Burg (1965) found that resealing the calyx scar of harvested cantaloupe melons with wax reduced their respiration rate by 10%. This suggests that the limited gas permeance of fruit tissues may be one factor restricting their respiration rate. Harvesting the fruit, and thereby increasing permeance, may facilitate a larger respiratory climacteric during ripening than would otherwise be possible.

A number of studies have suggested that differences between ripening behaviour on and off the

plant are due to 'plant factors'. In their study on the ripening of apples, Sfakiotakis and Dilley (1973) showed that a substance produced by the leaves and translocated to the fruit via the phloem inhibited ripening. The production of different amounts of these factors may account for variations in ripening physiology seen among cultivars of the same species. Different amounts of plant factors due to difference in plant vigour could also be responsible for the variations in ripening behaviour found in this study.

Together, the results of these experiments suggest that the respiratory climacteric may not be essential for ripening in climacteric fruit. Increases in respiration rate do not appear to be required to fuel the biochemical changes that occur during ripening. Rather, it is the necessary accompaniment of ethylene action in detached fruit. This confirms the proposal of a number of authors that it is changes in ethylene production that characterise climacteric fruit (McGlasson et al., 1978), the respiratory climacteric itself being primarily an artefact of harvest.

References

- Abdi, N., Holford, P., McGlasson, W.B., Mizrahi, Y., 1997. Ripening behaviour and responses to propylene in four cultivars of Japanese plums. *Postharvest Biol. Technol.* 12, 21–34.
- Abdi, N., McGlasson, W.B., Holford, P., Williams, M., Mizrahi, Y., 1998. Responses of climacteric and suppressed-climacteric plums to treatment with propylene and 1-methylcyclopropene. *Postharvest Biol. Technol.* 14, 29–39.
- Abeles, F.B., Morgan, P.W., Saltveit, M.E. Jr., 1992. *Ethylene in Plant Biology*. Academic Press, San Diego.
- Adato, I., Gazit, S., 1974. Postharvest response of avocado fruits of different maturity to delayed ethylene treatments. *Plant Physiol.* 53, 899–902.
- Andrews, J., 1995. The climacteric respiration rise in attached and detached tomato fruit. *Postharvest Biol. Technol.* 6, 287–292.
- Ayub, R., Guis, M., Ben Amor, M., Gillot, L., Roustan, J.P., Latche, A., Bouzayen, M., Pech, J.C.P., 1996. Expression of ACC oxidase antisense gene inhibits ripening of cantaloupe melon fruits. *Nat. Biotechnol.* 14, 862–866.
- Ben-Yehoshua, S., Robertson, R.N., Biale, J.B., 1963. Respiration and internal atmosphere of avocado fruit. *Plant Physiol.* 38, 194–201.

- Bernadac, A., Jean-Baptiste, I., Bertoni, G., Morard, P., 1996. Changes in calcium contents during melon (*Cucumis melo* L.) fruit development. *Sci. Hort.* 66, 181–189.
- Biale, J.B., Young, R.E., 1981. Respiration and ripening in fruits-retrospective and prospect. In: Friend, J., Rhodes, M.J.C. (Eds.), *Recent advances in the biochemistry of fruits and vegetables*. Academic Press, New York, pp. 1–39.
- Blanpied, G.D., 1993. Studies of the ‘tree factor’ that inhibits the ripening of attached apples. *Acta Hort.* 343, 6–11.
- Bliss, M.L., Pratt, H.K., 1979. Effect of ethylene, maturity, and attachment to the parent plant on production of volatile compounds by muskmelons. *J. Am. Soc. Hort. Sci.* 104, 273–277.
- Boller, T., 1990. Ethylene and plant–pathogen interactions. In: Flores, H.E., Arteca, J.C., Shannon, J.C. (Eds.), *Polyamines and Ethylene: Biochemistry, Physiology, and Interactions*. American Society of Plant Physiologists, Rockville, pp. 138–145.
- Bower, J.H., Jobling, J.J., Patterson, B.D., Ryan, D.J., 1998. A method for measuring the respiration rate and respiratory quotient of detached plant tissues. *Postharvest Biol. Technol.* 13, 263–270.
- Bower, J., Patterson, B.D., Jobling, J.J., 2000. Permeance to oxygen of detached *Capsicum annuum* fruit. *Aust. J. Exp. Agric.* 40, 457–463.
- Burg, S.P., Burg, E.A., 1962. Role of ethylene in fruit ripening. *Plant Physiol.* 37, 179–189.
- Burg, S.P., Burg, E.A., 1965. Gas exchange in fruits. *Physiol. Plant* 18, 870–884.
- Cameron, A.C., Yang, S.-F., 1982. A simple method for the determination of resistance to gas diffusion in plant organs. *Plant Physiol.* 70, 21–23.
- Dostal, H.C., Leopold, A.C., 1967. Gibberellin delays ripening in tomatoes. *Science* 158, 1579–1580.
- Elad, Y., 1990. Production of ethylene by tissues of tomato, pepper, French bean and cucumber in response to infection by *Botrytis cinerea*. *Physiol. Mol. Plant Pathol.* 36, 277–287.
- Golding, J.B., Shearer, D., Wyllie, S.G., McGlasson, W.B., 1998. Application of 1-MCP and propylene to identify ethylene-dependent ripening processes in mature banana fruit. *Postharvest Biol. Technol.* 14, 87–98.
- Guis, M., Botondi, R., Ben Amor, M., Ayub, R., Bouzayen, M., Pech, J.C., Latché, A., 1997. Ripening-associated biochemical traits of cantaloupe charentais melons expressing an antisense ACC oxidase transgene. *J. Am. Soc. Hort. Sci.* 122, 748–751.
- Hadfield, K.A., Rose, J.K.C., Bennett, A.B., 1995. The respiratory climacteric is present in Charentais (*Cucumis melo* cv. Reticulatus F1 Alpha) melons ripened on or off the plant. *J. Exp. Bot.* 46, 1923–1925.
- Jiang, Y., Joyce, D., Macnish, A.J., 1999. Extension of the shelf life of banana fruit by 1-methylcyclopropene in combination with polyethylene bags. *Postharvest Biol. Technol.* 16, 187–193.
- Knee, M., 1995. Do tomatoes on the plant behave as climacteric fruits? *Physiol. Plant.* 95, 211–216.
- Lacan, D., Baccou, J.C., 1996. Changes in lipids and electrolyte leakage during non-netted muskmelon ripening. *J. Am. Soc. Hort. Sci.* 121, 554–558.
- Laties, G.G., 1995. Franklin Kidd, Charles West and F.F. Blackman: the start of modern postharvest physiology. *Postharvest Biol. Technol.* 5, 1–10.
- Lyons, J.M., McGlasson, W.B., Pratt, H.K., 1962. Ethylene production, respiration and internal gas concentrations in cantaloupe fruits at various stages of maturity. *Plant Physiol.* 37, 31–36.
- McGlasson, W.B., 1970. The ethylene factor. In: Hulme, A.C. (Ed.), *The Biochemistry of Fruits and their Products*, vol. 1. Academic Press, London and New York, pp. 475–519.
- McGlasson, W.B., Wade, N.L., Adato, I., 1978. Phytohormones and fruit ripening. In: Lethan, S., Goodwin, P.B., Higgins, T.J. (Eds.), *Plant Hormones and Related Compounds—A Comprehensive Treatise*, vol. 2. Elsevier, Amsterdam, pp. 447–493.
- Mead, R., Curnow, R.N., Hasted, A.M., 1993. *Statistical Methods in Agriculture and Experimental Biology*, second ed. Chapman and Hall, London.
- Mullins, E.D., McCollum, T.G., McDonald, R.E., 2000. Consequences on ethylene metabolism of inactivating the ethylene receptor sites in diseased non-climacteric fruit. *Postharvest Biol. Technol.* 19, 155–164.
- Pooter, H.L., de Schamp, N.M., de Pooter, H.L., 1989. Involvement of lipoxygenase-mediated lipid catabolism in the start of the autocatalytic ethylene production by apples (cv. Golden Delicious): a ripening hypothesis. *Acta Hort.* 258, 47–53.
- Pratt, H.K., Goeschl, J.D., 1969. Physiological roles of ethylene in plants. *Annu. Rev. Plant Physiol.* 20, 541–584.
- Pratt, H.K., Goeschl, J.D., Martin, F.W., 1977. Fruit growth and development, ripening, and the role of ethylene in the ‘Honey Dew’ muskmelon. *J. Am. Soc. Hort. Sci.* 102, 203–210.
- Rogiers, S.Y., Knowles, N.R., 1999. A comparison of pre-harvest and postharvest ethylene production and respiration rates of saskatoon (*Amelanchier alnifolia* Nutt.) fruit during development. *Can. J. Bot.* 77, 323–332.
- Romani, R.J., 1987. Senescence and homeostasis in postharvest research. *Hortic. Sci.* 22, 865–868.
- Saltveit, M.E. Jr., 1993. Internal carbon dioxide and ethylene levels in ripening tomato fruit attached to or detached from the plant. *Physiol. Plant* 89, 204–210.
- Sfakiotakis, E.M., Dilley, D.R., 1973. Internal ethylene concentrations in apple fruits attached to or detached from the tree. *J. Am. Soc. Hort. Sci.* 98, 501–503.
- Shellie, K.C., Saltveit, M.E. Jr., 1993. The lack of a respiratory rise in muskmelon fruit ripening on the plant challenges the definition of climacteric behaviour. *J. Exp. Bot.* 44, 1403–1406.

- Shiomi, S., Wamocho, L.S., Agong, S.G., 1996. Ripening characteristics of purple passion fruit on and off the vine. *Postharvest Biol. Technol.* 7, 161–170.
- Smock, R.M., 1972. Influence of detachment from the tree on the respiration of apples. *J. Am. Soc. Hortic. Sci.* 97, 509–511.
- Wang, C.Y., Mellenthin, W.M., Hansen, E., 1972. Maturation of 'Anjou' pears in relation to chemical composition and reaction to ethylene. *J. Am. Soc. Hortic. Sci.* 97, 9–12.
- Yang, S.-F., Liu, Y., Lau, O.L., 1986. Regulation of ethylene biosynthesis in ripening apple fruits. *Acta Hortic.* 179, 711–720.