

Short Communication

Effect of Ethylene on the Permeability of Excised Cantaloupe Fruit Tissue¹

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Changes in tissue permeability are associated with the maturation and ripening of fruits. Blackman and Parija (3) suggested that permeability changes may cause the climacteric rise in respiration which is a characteristic of ripening in most fruits. Sacher (13) showed that leakage of solutes increased progressively with the climacteric rise in ripening banana and avocado fruits, and he noted that these results were consistent with the hypothesis that alteration in membrane properties may cause the climacteric. Ben-Yehoshua (2) reported that leakage of cellular materials from excised disks of avocado fruits began before the onset of the climacteric and continued to increase during the rise in respiration. He agreed with Sacher that changes in membrane permeability may bear a causal relationship to the climacteric. Baur and Workman (1) noted that changes in permeability and the onset of the climacteric apparently occurred simultaneously in banana fruits. Burg et al. (5) concluded that solute efflux from banana tissue was simply a reflection of sugar content, and that permeability did not change with ripening. However, using solute uptake to estimate free space, Sacher (14) has shown conclusively that permeability does increase with ripening of banana fruits and, indeed, that changes in permeability precede the onset of the climacteric rise.

There is evidence that an increase in ethylene production also precedes the climacteric rise in respiration. This has been shown in bananas (4) and in Honey Dew melons (12). Recalculation of the data of Lyons et al. (7) indicates that an increase in internal ethylene concentration probably precedes the onset of the climacteric in cantaloupe fruits, also.

Von Guttenberg and Beythien (6) and others have suggested that ethylene increases the permeability of plant tissue, and Lyons and Pratt (8) have shown that ethylene increased the permeability of isolated mitochondria. However, Burg et al.

(5) reported that ethylene did not alter the pattern of leakage from pea sections and argued that permeability of the tonoplast remained unaltered in banana fruits despite the presence of ethylene.

The present report concerns the effect of a short-term ethylene treatment on the water flux of excised tissue of cantaloupe fruits, in relation to the failure of the same treatment to influence the climacteric rise in respiration.

Cantaloupe fruits (*Cucumis melo* L., var. *reticulatus* Naud., cv. Powdery Mildew Resistant No. 45) were field grown. Growth patterns, ethylene production, respiration, and the ripening and respiration response to exogenous ethylene have been described for fruits of this cultivar (7, 9, 10, 11). Results reported here are from 2 separate experiments, one using fruits 15 days from anthesis and the second using fruits 30 days from anthesis. Administration of ethylene (100 ppm in a continuous air stream) and measurement of respiration (CO₂ evolution) were conducted in a 20° room by methods standard in this laboratory (10). Ethylene treatments were applied in 2 distinct situations: A) tissue disks were excised, and ethylene was applied to them only after excision; B) ethylene was applied to intact fruits, and the experimental tissue disks were excised after the ethylene treatment was terminated. Excision of tissue and the subsequent incubation and weighings were done in a 25° room. Each experiment was repeated with insignificant differences in procedure and results.

Twelve fruits of the same age (either 15 or 30 days from anthesis) were harvested and surface sterilized. From locations spaced around the equatorial areas of 2 of the fruits, 10 pairs of tissue plugs 12 mm in diameter were cut radially through the fruit wall. The excisions for each pair of plugs were located so that the inner ends of the 2 plugs were impinging. The inner 2 mm of tissue of each plug was discarded, and the next 2 mm excised as an experimental disk. Two paired samples of 10 disks each were prepared in this way, 1 disk to each sample from each pair of plugs. The disks were rinsed, blotted and weighed, and each of the 2 samples was placed in 100 ml of deionized water. They were moved to a 20° room,

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where humidified air was bubbled through one sample and a mixture of humidified air and ethylene (100 ppm) through the other. After 1 hour of this treatment, the samples were returned to the 25° room, weighed, and each sample placed in 250 ml of deionized water through which filtered humidified air was slowly bubbled. The samples were weighed at 1-hour intervals and placed in fresh deionized water after each weighing. Conductivity of the ambient solutions was measured after each weighing, and the dry weight of each sample was determined at the end of the incubation period. Figure 1-A illustrates the course of weight change of the ethylene-treated and of control disks from fruits of 2 ages.

The remaining fruits were kept at 20° for 48 hours after harvest and then were placed in 2 respiration chambers, 5 fruits in each. For 3 hours, a continuous flow of air was passed through 1 chamber, and a mixture of air and ethylene (100 ppm) through the other. Two ethylene-treated fruits and 2 control fruits were then removed from the chambers to the 25° room, and 10 radial plugs were taken from the equatorial region of each. Disks were excised as previously described. Each sample of 20 disks was weighed and placed in 400 ml of aerated deionized water. At 1-hour intervals the samples were weighed and placed in fresh deionized water. Weight changes are shown in figure 1-B. Each of the remaining 6 fruits was placed in an individual respiration jar, and CO₂ evolution was measured at 24-hour intervals. Fig-

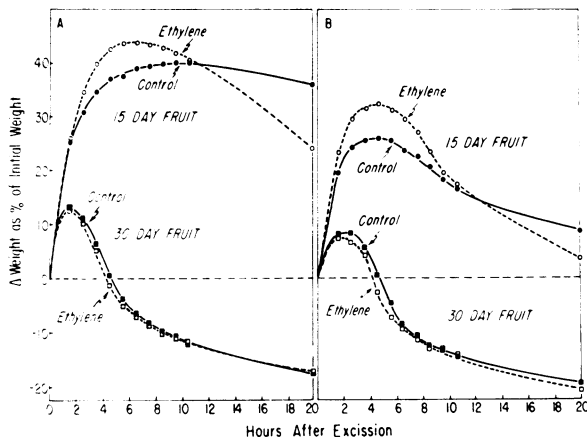


FIG. 1. Water flux expressed as change in weight of excised tissue disks incubated in deionized water at 25° for 20 hours after excision from cantaloupe fruits of 2 ages. Solid Line: Control. Broken line: Ethylene treated. A) Ethylene applied (100 ppm, 1 hr) to the disks after excision. Average weight change of a sample of 10 disks. B) Ethylene (100 ppm) applied to intact fruits for the 3 hours preceding excision of the disks. Average weight change of a sample of 20 disks. To correspond to the fruits used for measurement of respiration, these fruits were held for 48 hours after harvest before ethylene was applied.

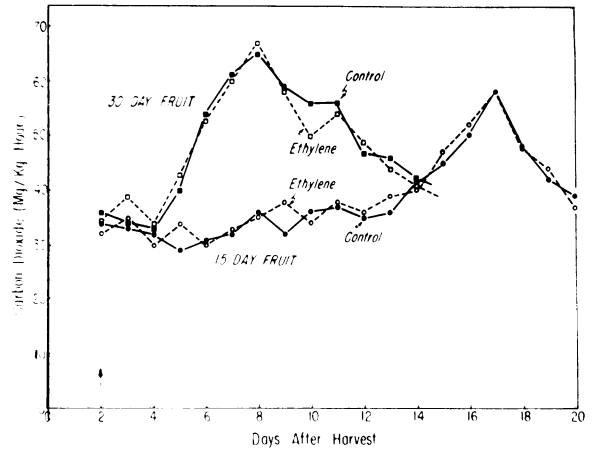


FIG. 2. Respiration rates of intact cantaloupe fruits of 2 ages, expressed as mg. CO₂ evolved per kilogram of fresh weight per hour at 20°. Averages of 3 fruits. Ethylene (100 ppm) was applied for 3 hours at the time indicated by arrow. Fruits harvested 15 days after anthesis: ●, control; ○, ethylene treated. Fruits harvested 30 days after anthesis: ■, control; □, ethylene treated.

ure 2 shows the course of respiration of control and of ethylene-treated fruits through the climacteric rise.

The weight change of the tissue disks in deionized water was predominantly a change in water content of the tissue. The fraction of weight loss due to solute efflux from tissue of young fruit was negligible, except for its influence upon osmolarity within the tissue. Ethylene treatment of immature (15-day) fruit, whether applied to excised tissue (fig 1-A) or to the intact fruit (fig 1-B), caused an increase in water influx, followed by strongly accelerated efflux. In 30-day fruit, in contrast, the ethylene treatment had no detectable influence upon either water influx or efflux. In tissue from fruit of this age, the pattern of water flux is changed greatly from that of 15-day fruit. Influx is much reduced, and efflux has become predominant. The intact 30-day fruit is capable of a well-defined climacteric without additional stimulation by exogenous ethylene (10), and it is probable that native ethylene has already exerted a large influence.

In contrast to the effect of water flux in tissue of young fruit (fig 1-B), it is shown in figure 2 that the short ethylene treatment (3 hrs, 100 ppm) was not sufficient to accelerate the climacteric or otherwise alter the gross respiration pattern in either 15- or 30-day intact fruits. It is known, however, that a 6-hour treatment with 100 ppm of ethylene causes a rise in respiration in 10- to 30-day fruits of this cultivar, and if the treatment is prolonged to 24 hours the climacteric is accelerated and the magnitude of the respiratory peak is increased (10).

The initial ethylene-induced increase in water influx in tissue of 15-day fruits might result from metabolically increased osmolarity, from increased wall elasticity or plasticity, or from increased permeability. Our argument for increased permeability is based upon the accelerated efflux which follows the initial influx. Increased wall elasticity cannot be the cause of efflux. An increase in osmolarity might result in an increased efflux of solute, not necessarily requiring increased permeability, and this would lead to a declining rate of water influx and then to water efflux. Once osmolarity had declined to the level of the control, however, only increased permeability could account for efflux exceeding that of the control. The marked acceleration of efflux from tissue of 15-day fruits in response to the short ethylene treatment is clearly shown in figure 1-A, B.

We suggest, therefore, that exogenous ethylene may alter the pattern of water flux in tissue of young cantaloupe fruits, and that the change reflects increased permeability. Moreover, since a change in permeability was induced by ethylene treatment too brief to cause the typical respiratory response to ethylene, it is apparent that the permeability response must precede the respiratory response during continuous exposure to ethylene. Although it is also manifest that the change in permeability induced in these experiments did not cause a detectable acceleration of the climacteric, the results do not refute the hypothesis that increased permeability is a cause of the climacteric. Pre-climacteric changes in compartmentation may be complex and sequential. It remains to be shown whether permeability bears a direct causal relationship to the various parameters of ripening.

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