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Respiration and Ethylene Production in Fruits of Species and Cultivars of *Psidium* and Species of *Eugenia*[1]

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Abstract. **Fruits of 4 species and cultivars of guava (*Psidium guajava* L. cv. Beaumont, *P. cattleianum* Sabine, *P. cattleianum* f. *lucidum* Degener, and *P. guajava* L. cv. Allahabed Safeda) were found to be climacteric in their respiratory behavior with C_2H_4 triggering the respiratory rise. Fruits of 4 species of *Eugenia* (*Eugenia malaccensis* L., *E. cumini* (L.) Druce, *E. uniflora* L., and *E. jambos* L.) exhibited typical respiratory behavior of non-climacteric fruits including the response to exogenous C_2H_4 treatments. The possibility of using respiratory data as a physiological tool for taxonomic differentiation of plants is discussed.**

Contradictory reports exist as to the respiratory pattern of the fruit of guava, i.e. some indicated that it is climacteric (2) and others non-climacteric (5). We report here the differences in respiration and C_2H_4 production between the fruits of species and cultivars of *Psidium* and *Eugenia* (Myrtaceae).

Materials and Methods

Fruits of the following 4 guava species and cultivars were used in these investigations: *Psidium guajava* L. cv. Beaumont (Beaumont guava), *P. cattleianum* Sabine (purple strawberry guava, Cattley guava), *P. cattleianum* f. *lucidum* Degener (yellow strawberry guava, yellow Cattley guava), and *P. guajava* L. cv. Allahabed Safeda (white guava). Fruits of the following 4 species of *Eugenia* were also used: *Eugenia malaccensis* L. (mountain apple, Malay apple), *E. cumini* (L.) Druce (Java plum, jambolan plum), *E. uniflora* L. (Surinam cherry, pitanga), and *E. jambos* L. (rose apple). Fruits were obtained from trees at the Hawaii Agricultural Experiment Station, Lyon Arboretum of the University of Hawaii, or private domiciles. At anthesis the blossoms were enclosed in bags made of double layered cheesecloth to prevent attack by the oriental fruit fly (*Dacus dorsalis* Hendel). Covering did not interfere with fruit development.

The guava fruits for investigations on respiration (CO_2 production) and C_2H_4 production were picked at the stage when the fruit surface color had turned light green from dark green. This stage was assumed to be the time when maximum growth (mature green stage) of the fruit had been attained. The mountain apple fruits were picked at the light green stage (mature green) and red stage (ripe), and the rose apple fruits were also picked at the mature green stage (light green) and the ripe stage (yellow for respiration and C_2H_4 studies. The surface color of Surinam cherry changes from green to yellow to orange to red as the fruit grows and ripens, and measurements of diameter (middle of fruit) and length (blossom end to stem end) of the fruit indicated that the fruit reaches its maximum growth when the surface color is light orange (mature green stage). Fruits of this color to red (ripe) stage were used for respiration and C_2H_4 studies. Similar measurements indicated that Java plum fruits attain their maximum growth at the light purple stage (the fruit surface color changes from green to red to purple as the fruit grows and ripens). Light purple-colored (mature green stage) as well as red-colored (immature stage) fruits were used in these investigations.

Within 2 hr after picking, the fruits were installed in respiration experiments. They were individually held in air-tight glass jars (144 or 497 ml) in the laboratory (24.0 – 25.7°C; 64 – 70% relative humidity) for CO_2 and C_2H_4 determinations. After 1 hr, duplicate samples (1 ml each) of the atmosphere surrounding the fruit were withdrawn with a gastight syringe for CO_2 analysis with a gas chromatograph (Varian Aerograph 90-F) equipped with a thermal conductivity detector unit, silica gel column (0.6 x 35.6 cm), and He_2 carrier gas. One hour later, similar duplicate samples were withdrawn from the same jars for C_2H_4 analysis by gas chromatography (Aerograph Hy-Fi 600-D equipped with a hydrogen flame ionization unit, alumina column (0.3 x 182.9 cm), and N_2 carrier gas). Sampling was done daily until the fruits deteriorated. Between samplings the fruits were subjected to a moving stream of humidified air in the respiration jars at the rate of 50 ml per hr.

Emanations of some fruits were bubbled into $Hg(ClO_3)_2$ solution (0.25 M red mercuric oxide in 2.0 M perchloric acid) for 18-24 hr (13). The trapped gas after being liberated with 4.0 N LiCl was analyzed for C_2H_4 by gas chromatography. Some fruits were treated with C_2H_4 in air-tight glass jars with varying amounts of pure C_2H_4 injected into the jars in the laboratory.

Results and Discussion

Guavas. The guava species and cultivars exhibited typical respiratory patterns of climacteric class of fruits with the increase in C_2H_4 production preceding the respiratory rise by 1 day (Fig. 1). Whereas C_2H_4 production increased immediately after 1 day, respiration decreased before commencing to rise as the fruit turned yellow (light yellow in white guava) and ripened. The climacteric nature of guava fruits was shown in these results which verified the findings of some researchers (2) and contradicted those of others (5) who probably were not able to obtain good quality fruits of proper maturity for their studies.

Rose apple. Respiration and C_2H_4 production of rose apple as shown in Table 1 indicate the non-climacteric nature of the species. Respiration and C_2H_4 production declined during the first 3 days, then remained more or less constant during the remainder of the storage period, and no significant fruit surface color change occurred. Respiration of fruits of varying stages of development from mature green to ripe immediately after picking also showed the non-climacteric nature of the species. The respiration of fruits subjected to 2,000 or 3,000 ppm C_2H_4 for 18 hr rose immediately after treatment, then declined to the level of the untreated fruit and did not peak. This is also a typical response of non-climacteric fruit to C_2H_4 treatment (3, 4, 6).

Mountain apple. Respiration of mountain apple also indicates the non-climacteric nature of this species with CO_2 production declining the first 3 days, then remaining more or less constant

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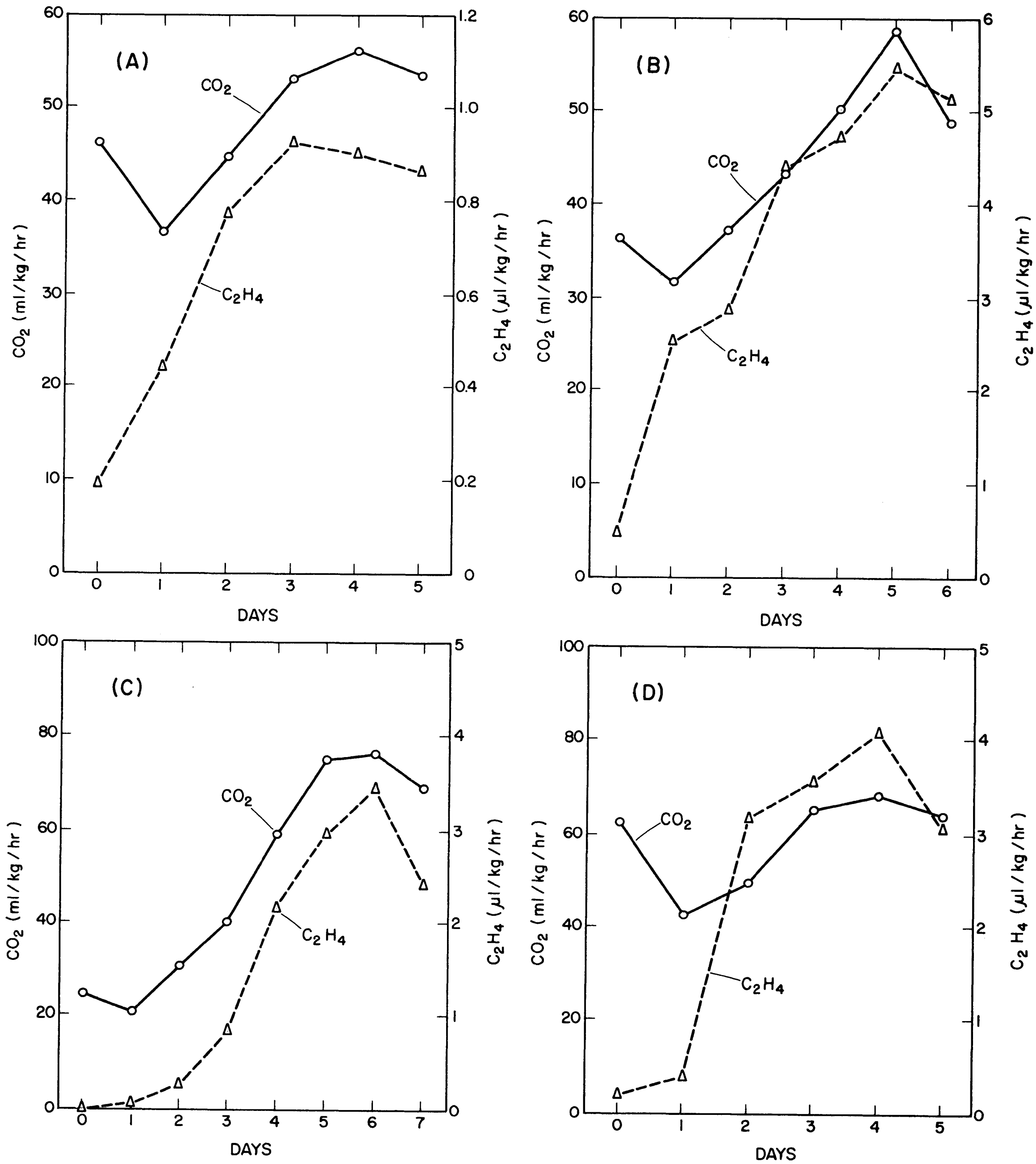

Fig. 1. Respiration (CO_2) and C_2H_4 production in mature green yellow strawberry guava (A), Beaumont guava (B), white guava (C), and purple strawberry guava (D).

during the remainder of the storage period with no detection of C_2H_4 production (Table 1). No significant fruit surface color change occurred during this period. Fruits of varying stages of development from mature green to ripe analyzed immediately after picking also indicated the non-climacteric nature of the species with no detection of C_2H_4 production. By the $Hg(ClO_3)_2$ procedure to trap C_2H_4 followed by analysis by gas chromatography, C_2H_4 (0.15 µl/kg·hr) was detected in the emanations of mountain apple. As in the case of rose apple, the respiration of mountain apple fruits subjected to 2,000 or 3,000 ppm C_2H_4 for 18 hr rose immediately after treatment, then declined to the level of the untreated fruits and did not peak. On the basis of results of only one experiment, the mountain apple was tentatively classified as intermediate between climacteric and non-climacteric class of fruits in respiratory behavior (5). We now place this fruit in the non-climacteric class.

Java plum. Although Java plum fruits of light purple stage (fully grown) exhibited non-climacteric respiratory patterns with no evidence of C_2H_4 production, comparison was made among fruits of various stages of surface color development (Table 2). In all cases, there was no evidence of climacteric respiratory pattern or any significant color change, and no C_2H_4 production was detected. However, about 26 ppm of C_2H_4 was trapped in the $Hg(ClO_3)_2$ solution after emanations of mature green fruit were exposed to the solution for 20 hr. Respiration of mature green fruit treated with 10 ppm C_2H_4 for 18 hr increased immediately after treatment, then decreased rapidly to the level of untreated fruit and continued to decrease gradually without peaking.

Surinam cherry. The respiratory pattern of mature green fruit of this species also indicates the non-climacteric nature with no detection of C_2H_4 by the standard procedure or any

Table 1. Respiration and C_2H_4 production in mature green rose apple and mountain apple fruits (average of 8 fruits per species) at 24.0 – 25.7°C.

Day	Rose apple CO_2 (ml/kg·hr)	Rose apple C_2H_4 (µl/kg·hr)	Mountain apple CO_2 (ml/kg·hr)	Mountain apple C_2H_4 (µl/kg·hr)
0	54.8	1.52	32.8	0
1	35.1	1.00	27.9	0
2	24.2	0.41	28.5	0
3	20.1	0.21	22.1	0
4	20.8	0.17	21.9	0
5	22.5	0.10	25.0	0
6	24.2	0.04	25.0	0
7	23.1	0.23	26.0	0
8	23.4	0.08	25.6	0

change in surface color of the fruits (Table 3). However, ripe fruits held for 22 hr with $Ca(OH)_2$ as a CO_2 scrubber produced C_2H_4 at the rate of 0.86 µl/kg·hr. When mature green fruits were treated with 25 ppm C_2H_4 for 22.5 hr, respiration increased immediately, then decreased to about the original level for the rest of the storage period, but this level was much higher than that of untreated fruit (Table 3). The most significant effect of C_2H_4 treatment was the change in surface color of the fruit from the initial light orange to red.

The fruits of the 4 species of *Eugenia* were thus found to be non-climacteric in their respiratory behavior as indicated by their lack of respiratory and C_2H_4 peaks and their typical response to C_2H_4 treatment. These non-climacteric species have one factor in common with the climacteric *Psidium* species and cultivars, and that is, they are all non-starchy fruits as indicated by the IKI test. The similarity in the respiratory behavior of fruits of different species and cultivars of the same genus *(Psidium* or *Eugenia)* and the lack of similarity between the 2 genera in the same family *(Myrtaceae)* were shown in these results. Chemical composition as a taxonomic tool for classification and differentiation of plants (chemotaxonomy) has been investigated (11, 12), especially in the closely related species of the genus *Citrus* (7, 9, 10). The fruit of a non-ripening mutant of tomato was found to be non-climacteric in contrast to the climacteric fruit of a normal tomato (8). This probably suggests the possibility of separating a cultivar into sub-cultivars based on respiratory behavior. Respiratory behavior of fruit as a possible physiological tool for taxonomic differentiation of plants may be suggested by these investigations. Although *Eugenia malaccensis, E. cumini,* and *E. jambos* are now placed in the genus *Syzygium* (family *Myrtaceae*) by some taxonomists (1), the suggested possibility of using respiratory behavior as a taxonomic tool may still have merit.

Table 2. Respiration of Java plum fruit of varying stages of development (average of 8 fruits per color stage) at 24.0 – 25.7°C.

Fruit color stage	CO_2 (ml/kg·hr) on day[z] 0	1	2	3	4	5	6	7	8
10-30% red	38.8	23.6	17.8	16.5	17.4	19.8	17.0	13.2	14.3
40-70% red	36.3	20.9	17.6	15.6	16.1	17.3	16.7	14.3	11.4
80-100% red	33.6	18.9	16.8	17.0	16.9	15.4	11.6	10.8	11.3
5-10% purple[y]	34.4	29.6	23.2	22.0	18.0	17.1	14.0	10.2	
80-95% purple	27.9	19.0	16.0	14.9	12.0				

[z]No C_2H_4 detected.
[y]Fully grown stage.

Table 3. Effect of C_2H_4 on respiration and color development in mature green Surinam cherry fruit (average of 10 fruits per treatment) at 24.0 – 25.7°C.

	Treatment							
	+ C_2H_4[z]				Control			
Day	CO_2 (ml/kg·hr)	% light orange fruits	% orange fruits	% red fruits	CO_2 (ml/kg·hr)	% light orange fruits	% orange fruits	% red fruits
0	18.7	100	0	0	18.6	100	0	0
1	49.1	0	100	0	22.7	100	0	0
2	21.5	0	100	0	15.3	100	0	0
3	20.6	0	100	0	14.0	100	0	0
4	21.0	0	90	10	14.9	100	0	0
5	22.2	0	20	80	12.1	100	0	0
6	21.2	0	20	80	12.4	100	0	0
7	22.1	0	10	90	12.1	100	0	0
8	20.5	0	0	100	9.7	100	0	0

[z]25 ppm for 22.5 hr.

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Honey Bee Foraging and Resultant Seed Set among Male-fertile and Cytoplasmically Male-sterile Carrot Inbreds and Hybrid Seed Parents[1,2]

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Abstract. **A study of foraging by honey bees (*Apis mellifera* L.) among cytoplasmically male-sterile and male-fertile seed parents of carrot (*Daucus carota* L.) revealed that honey bee discrimination between the fidelity to carrot phenotype and genotype were evident and often extreme. Some lines were extensively visited while others were virtually ignored. Wide differences in seed set were evident among male-sterile F_1's and inbreds and male-fertile lines. Differences in seed yield were correlated with foraging preferences, but the quality of nectar from the stomachs of bees was not.**

A complex of problems ranging from nonrandom pollinator foraging activity to genetic incompatibility between seed parents has resulted in poor commercial production of hybrid seed from carrots. Observation has shown that various cytoplasmically male-sterile and male-fertile carrot lines bloom at different times and are probably differentially attractive to pollinators even when they are blooming simultaneously. This floral variation among maternal and paternal lines inhibits row crossing by honey bees, the principal pollinator of commercially produced carrots (1).

Honey bees, whether they are foraging for nectar or for pollen, readily discriminate between nectar and pollen sources on the basis of foraging stimuli (e.g. aroma, taste, color, size, and shape) and retain a marked fidelity to a single source (2, 3, 4). Apiculturists familiar with this behavior ordinarily think of discrimination as limiting interspecific foraging activity. However, such extreme differences are evident in the floral characteristics of selected parental lines of carrots that we must recognize the great potential for intravarietal discrimination by bees.

Since honey bees seemed to forage discriminately between carrot genotypes and some were rarely visited by bees and as a result set seed poorly, we compared levels of foraging activity between and the resultant seed set among selected male-sterile and male-fertile genotypes.

Methods and Materials

The studies were conducted from 1973 to 1976 in field isolation cages at Madison, Wisconsin. Initially we ascertained only whether bees discriminated between corolla color, since male-fertile maintainer (M) lines are white, and male-sterile (S) lines are either off-white or green. In later studies we evaluated foraging based on genotype and then compared seed yields.

Foraging. 1973. Two identical 3-row screened plots were studied. In each a center row (7.6 m) containing white corolla male-fertile (M) plants all of the same line was flanked

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