Table 2. In-vitro micropropagation of 'Cohen' citrange, media sequence.

1. Ovule culture:	EME medium MT Basal + 0.5 g/liter malt extract, and 50 g/liter sucrose
2. Embryo proliferation:	EME medium
3. Embryo enlargement:	1500 medium MT Basal + 1.5 g/liter malt extract, and 50 g/liter sucrose
4. Embryo germination:	B+ medium MT Basal + 0.02 mg/liter NAA, 1.0 mg/liter GA_3, 10 mg/liter coumarin, and 25 g/liter sucrose
5. Shoot multiplication:	DBA3 medium MT Basal + 3 mg/liter BA, 0.02 mg/liter 2,4-D, and 25 g/l sucrose
6. Rooting:	RMAN medium 1/2 strength MT Basal + 0.02 mg/liter NAA, 0.5 g/liter activated charcoal, and 25 g/liter sucrose

adequate commercial evaluation, and the methods described should be amenable to the propagation of any other citrange of interest where adequate plant numbers are difficult to obtain from seed.

Inadequate nucellar polyembryony no longer has to be a limiting factor in selecting germplasm for citrus rootstock evaluation. Many genotypes are amenable to alternative methods of propagation, and with minimal research, these methods could be made economically competitive. Examination of a broader germplasm base (including wide interspecific and intergeneric hybrids being produced by citrus improvement programs) should accelerate the identification of improved rootstock genotypes that have the necessary disease/nematode resistance and stress tolerance, as well as adequate horticultural performance.

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RESPIRATION DIFFERENCES OF CITRUS ROOTSTOCK FEEDER ROOTS

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Additional index words. oxygen uptake, CO_2 evolution.

Abstract. **Oxygen uptake of root tips of 'Hamlin' orange (*Citrus sinensis* [L.] Osbeck) trees on 5 rootstocks grown in pots in a greenhouse was measured at 15 min intervals for 60 min. Roots of Sun Chu Sha mandarin (*C. reticulata* Blanco) took up more oxygen than those of rough lemon (*C. limon* Burm f.) and Cleopatra mandarin (*C. reticulata*). Carrizo citrange (*Poncirus trifoliata* [L.] Raf. × *C. sinensis*) and sour orange (*C. aurantium*) roots absorbed the least O_2. Carbon dioxide evolution of feeder roots collected from 'Valencia' orange (*C. sinensis*) trees on 12 rootstocks at 2 locations was measured by shaking 15.0 g of roots in calibrated flasks overnight. The flasks contained 100 ml of Hoagland's solution, and CO_2 was determined by gas chromatography of the headspace air. Roots of citrumelos (*C. paradisi* Macf. × *P. trifoliata*) from both locations produced the most CO_2 per square meter of root surface per hour. Sour orange, Carrizo citrange, and Volkamer lemon evolved the least CO_2.**

Root respiration is an important aspect of plant/soil relations. Lack of oxygen in the root zone has a wide range of detrimental effects on the above-ground parts of plants. The CO_2 liberated modifies the soil around the roots, and the energy released by respiration drives vital functions such as nutrient absorption. We investigated both components of respiration, the oxygen uptake of feeder roots of greenhouse-grown plants on 5 rootstocks and the CO_2 evolution of feeder roots of 12 citrus rootstocks of mature, producing trees in the field. In all cases, we used grafted plants because work with rootstock seedlings gives dubious results because of the strong influence of the scion on rootstock behavior (Smith, 1975).

Materials and Methods

Root tip samples 1-cm long (62-79 mg dry weight) were taken in October 1989 from 1-year-old 'Hamlin' orange (*Citrus sinensis* [L.] Osbeck) trees, 3 trees each on rough lemon (*C. limon* Burm. f.), sour orange (*C. aurantium* L.), Cleopatra mandarin, Sun Chu Sha mandarin (*C. reticulata* Blanco), and Carrizo citrange (*Poncirus trifoliata* [L.] Raf. × *C. sinensis*) rootstocks growing in 2.6-liter pots, in 60%

The authors are grateful to Mr. Orie Lee for making trees available for the field part of the experiment. Mention of a trademark, warranty, proprietary product, or vendor does not constitute a guarantee by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products or vendors that may also be suitable.

sand, 30% peatmoss, and 10% perlite medium. The roots were washed in tap water, blotted dry, and oxygen uptake of 3.0 g fresh weight samples was measured immediately with a Gilson differential respirometer (Gilson Medical Electronics, Middleton, WI). Oxygen uptake 15, 30, 45, and 60 min after insertion of the samples was expressed as µl O_2/mg dry weight.

To measure CO_2 evolution, parts of the feeder root system of 5 trees on each rootstock were collected from 0-20 cm depth with a spade under the trees of 12 rootstock tests at 2 locations near St. Cloud, FL. The samples, consisting of branch roots up to 2-mm thick with attached fine roots, were taken between 12:00 noon and 3:00 PM on April 4, 12, 17, and 18, 1990, with surrounding soil and immediately placed in ice chests for transport to the laboratory. There the roots were washed in a stream of tap water, blotted dry, and 15.0 g of feeder roots were inserted into calibrated 500-ml flasks containing 100 ml of Hoagland's solution. The flasks were sealed with rubber septa and shaken for 14 hr on a platform shaker at 24°C room temperature. A 1-ml headspace air sample from each flask was analyzed for CO_2 by standard gas chromatographic methods. Length and width of the roots were measured on 1.0 g subsamples with a Delta-T Rootlength Measurement System (Delta-T Devices, Ltd., Burwell, Cambridge, England), and root surface area was calculated from the data. Carbon dioxide evolution was expressed as $ml \cdot m^{-2}$ root surface · hr.

The trees at Location 1 were 16-year-old 'Valencia' orange (*C. sinensis*) on 11 rootstocks: 3 citrumelos (*C. paradisi* Macf. × *P. trifoliata*), F-80-8, F-80-3, and Swingle; the rough lemon variant Milam, rough lemon and Volkamer lemon (*C. limon* Burm. f.); a Rangpur × Troyer hybrid (*C. reticulata* hyb. × [*C. sinensis* × *P. trifoliata*]); Cleopatra mandarin; sweet lime (*C. aurantifolia* L.); sour orange; and Carrizo citrange. The sandy soil was a mixture of Pomello (sandy, siliceous, hyperthermic arenic haplohumods) and Blanton soils (loamy, siliceous, thermic grossarenic paleudults) modified by draining and mixing. Tree spacing was 4.5 × 7.5 m.

At Location 2, 12-year-old 'Valencia' trees were on 7 of the same rootstocks as in Location 1 (Swingle citrumelo, Cleopatra mandarin, sweet lime, Carrizo citrange, Rangpur × Troyer, Volkamer lemon, sour orange), and 1 additional rootstock, English Small Flowered trifoliate orange (*P. trifoliata*). They were planted 4.5 × 6.6 m on land created by dredging and mixing several soils, all of them sandy.

Results and Discussion

The oxygen consumption of the root tips grown in pots (Fig. 1) falls into 3 groups: 1) Sun Chu Sha, an old small-fruited Chinese mandarin recently released as a rootstock, absorbed the most O_2; 2) rough lemon and Cleopatra were intermediate; and 3) Carrizo and sour orange absorbed the least O_2. There is nothing in the performance characteristics of these rootstocks that could be correlated with the differences in respiration.

Carbon dioxide evolution of the excised field-grown feeder roots (Table 1) was highest for citrumelo roots and coincides reasonably well when the rootstocks at the 2 locations are compared (r = .69). English Small Flowered trifoliate orange, which was present only at Location 2, evolved CO_2 in the intermediate range between Swingle citrumelo and sour orange (Table 1). The relative respiration rates of Cleopatra, Carrizo, and sour orange in all 3 sets of root samples were similar, whether respiration was measured as oxygen consumption (Fig. 1) or CO_2 evolution (Table 1). Cultural practices at both field sites were very similar, except that Location 1 was irrigated by overhead gun while an under-the-tree sprinkler system was used at Location 2. Swingle citrumelo, currently the rootstock of 60% of citrus nursery stock in Florida, has mainly relatively unbranched, coarse feeder roots (Castle et al., 1988), and a high respiration rate (Table 1), a characteristic it seems to share with other grapefruit × trifoliate orange hybrids. Trees on Swingle citrumelo produce large crops on relatively small canopies (Castle et al., 1988; Wutscher and Bistline, 1988), but trees on citrumelo F-80-8 are not as productive (Wutscher and Bistline, 1988). Sour orange roots consistently had low respiration rates, yet trees on it are medium-sized and produce high-quality fruit (Castle et al., 1988). The experiment showed that feeder roots of different rootstock cultivars respire at varying rates, but the significance of this is not clear. It appears that feeder root respiration is not a good indicator of rootstock performance.

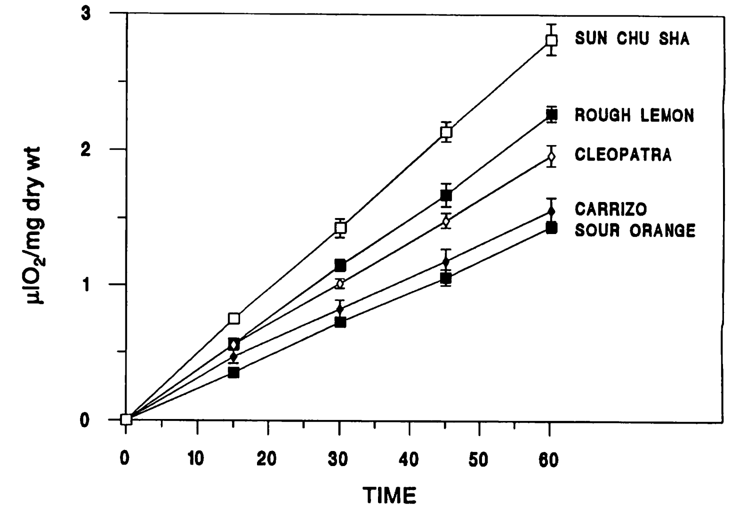

Fig. 1. Oxygen uptake of excised feeder root tips of 5 rootstock cultivars after 15, 30, 45, and 60 min in a respirometer. Bars show standard deviation; 3 replications each.

Table 1. Carbon dioxide evolution of citrus rootstock feeder roots (ml $CO_2 \cdot m^{-2}$ of root surface · hr.).

Location 1		Location 2	
Citrumelo F-80-8	707 a[z]		
Swingle citrumelo	598 b	Swingle citrumelo	731 a
Citrumelo F-80-3	573 b		
Milam	561 bc		
Rangpur × Troyer	521 bcd	Rangpur × Troyer	581 c
Volkameriana	520 bcd	Volkameriana	581 c
Cleopatra	512 bcd	Cleopatra	687 abc
Rough lemon	511 bcd		
Sweet lime	493 cd	Sweet lime	607 bc
Sour orange	491 cd	Sour orange 575 c	
Carrizo	438 d	Carrizo	598 c
		English Small Flowered trifoliate orange	634 bc

[z]Mean separation within columns by Duncan's multiple range test, p = 0.05.

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EFFECT OF SOIL TEMPERATURE AND FORCING METHOD ON SCION BUDBREAK AND GROWTH OF CITRUS NURSERY TREES

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Additional index words. budding, grafting, propagation.

Abstract. **Two experiments were conducted to determine the effect of soil temperature and forcing method on scion bud break and growth of Swingle citrumelo [*Citrus paradisi* (L.) Raf. × *Poncirus trifoliata* (L.) Osb.] and Cleopatra mandarin [*C. reticulata* Blanco] seedlings budded with 'Hamlin' orange [*C. sinensis* (L.) Osb.]. In experiment 1, budded Swingle (S) and Cleopatra (C) seedlings were grown at two soil temperatures (15 and 25°C) for seven weeks following bud forcing by bending. In experiment 2, scion buds were forced by: (1) cutting off the seedling top; (2) bending the seedling top over; or (3) bending + 6-benzylamino purine (BA) at 500 mg/l and grown at one of two soil temperatures (15 or 25°C). In experiment 1, the higher soil temperature treatment resulted in greater percent scion bud break, shorter period from forcing to bud break, and greater scion growth than the lower soil temperature treatment for S plants, but not for C plants. In experiment 2, cutting off rootstock tops resulted in greater percent bud break than bending at the low soil temperature. At the higher soil temperature, bending + BA resulted in 100% scion budbreak compared to bending which gave 75% budbreak. Forcing by bending + BA resulted in greater scion growth than cutting off rootstock tops.**

Inserted citrus scion buds are inconsistent in percent budbreak and time to budbreak following forcing (Halin et al., 1990; Maxwell and Lyons, 1979; Nauer et al., 1979; Orillos, 1953; Van der Poll, 1991). Possible factors influencing scion budbreak of citrus nursery trees include soil and air temperatures, photoperiod, forcing method and rootstock. Temperature is one of the most important environmental factors affecting callus growth needed for bud union formation following budding (Hartmann et al., 1990). Soil temperature has been shown to affect budbreak of sweet orange, sour orange, mandarin and rough lemon seedlings (Reuther, 1973). However, we know of no reports comparing the effects of soil temperature on scion budbreak of budded citrus nursery trees. Forcing method affects scion budbreak and growth. However, its influence depends on the rootstock and season of budding (prevailing environmental conditions) (Amih, 1980; Nauer and Boswell, 1981; Samson, 1986; Williamson et al., 1991). When cutting off is used to force scion growth, percent scion budbreak is often higher but scion growth is less compared to bending or lopping (Rouse, 1988; Williamson et al., 1992). Increased growth from lopping or bending was partially attributed to current rootstock top photosynthates translocated to the roots and scion (Williamson et al., 1992).

Swingle citrumelo is the most widely propagated citrus rootstock in Florida (Youtsey, personal communication, 1993). Scion budbreak is often low when bending or lopping is used to force scion bud growth on Swingle citrumelo. Therefore cutting off is commonly used with Swingle citrumelo but scion and root growth are probably less than would be achieved from lopping or bending. The objective of this study was to determine the effect of soil temperature and forcing method on budbreak and plant growth of 'Hamlin' orange budded on Swingle citrumelo rootstock.

Materials and Methods

Two experiments were conducted using 'Hamlin' orange budded on Swingle citrumelo and Cleopatra mandarin rootstocks. Swingle and Cleopatra seedlings were budded using the inverted T-bud procedure, wrapped for 3 weeks, and forced by bending the rootstock tops over and tying them in place. In experiment 2, bending and 2 additional forcing treatments were used: (1) cutting off the rootstock tops just above the scion bud; and (2) bending plus 6-benzylamino purine (BA) at 500 mg/l.

Experiment 1. Swingle and Cleopatra seedlings were transplanted to citripots (10.2 × 10.2 × 35.5 cm) containing a peat moss and perlite growing media (2:1 V:V) and grown in a greenhouse. About 4 months after transplanting, all plants were budded. Three weeks later, buds were unwrapped and forced by bending. After forcing, plants were immediately moved to a growth room. Plants were grown for 7 weeks at root temperatures of 15°C or 25°C using computer-controlled chest freezers accurate to ± 1°C. Plants were positioned so that the citripots were in the

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