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Promotion of Olive Fruit Abscission with 2-Chloroethyl-tris-(2-methoxyethoxy)-silane[1]

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Abstract. **Foliar applications of 2-chloroethyl-tris-(2-methoxy ethoxy)-silane (CGA 13586) caused abscission of olive fruits and, to much lesser extent, leaves due to its release of ethylene upon contact with water. Maximum reduction in fruit removal force occurred 7 to 8 days after application and seemed to be a direct effect on the abscission layer. The ethylene-releasing reaction rate was affected by the solution pH, increasing with higher pH values. Lower pH (ca. 6) gave greater abscission than did higher pH levels since the compound presumably hydrolyzed too rapidly in the latter for the ethylene released to cause abscission. CGA 13586 produced considerably higher initial ethylene release in comparisons with (2 chloroethyl)phosphonic acid (ethephon) but it was not as long lasting.**

An abscission-inducing chemical would be helpful in attaining adequate fruit removal, especially for small-fruited cultivars, in order to mechanize the table olive harvest in California by tree shaking. Fruits harvested for the "ripe" olive process are not completely mature and have a high attachment force. Furthermore, the fruits are mostly borne on long willowy shoots which do not transmit energy well from the shaker arm which is clamped to the trunk or to primary scaffold branches. The relatively small mass of the fruits also makes separation from the shoots difficult.

Efforts have continued over a number of years to find a suitable abscission-inducing agent, starting with tests in 1955 using maleic hydrazide (1). Ethephon (2) caused fruit abscission but also excessive leaf abscission. Excessively high leaf abscission in the fall reduces bloom the following spring (3). Cycloheximide (CHI) sprays caused (3) olive fruit abscission but excessive pitting of the fruits ruled out commercial use of this material on table olives.

In 1972 CIBA-GEIGY made an ethylene generating compound, 2-chloroethyl-tris-(2-methoxyethoxy)-silane (CGA 13586; 200 SCW formulation), available for trial (4). The tests reported here provide information concerning abscission effects of this material on olives.

Materials and Methods

Sprays of CGA 13586 were applied at 0, 500, 750, 1000, 1500, and 2000 ppm with a power sprayer on Oct. 24, 1972 to large fruiting limbs of 20 year old 'Manzanillo' olive trees. Regulaid (Colloidal Products Co., Petaluma, Ca.) at 0.3% was used as a surfactant in this and all subsequent spray applications. The weather was clear and sunny (24°C max; 13° min). Fruit removal force was measured with a dial push-pull gauge (Chatillon Model DPP-1 kg) adapted with a two-prong hook for attaching to olive fruits. Percentage leaf drop was obtained by vigorous hand shaking of sample shoots 13 and 28 days after spraying. Leaves were counted just before spraying and just after shaking.

Sprays of 4 concn of CGA 13586 were applied to entire trees of 'Mission', 'Manzanillo', and 'Sevillano' in the same orchard on Oct. 30, 1973. The weather was clear when the sprays were applied, with a maximum of about 18°C and minimum of about 10°. There was 22 mm of rain on the night of Nov. 6. Ten branch replicates were shaken on Nov. 8 with an air-powered limb shaker. Fruit and leaf removal was determined as described for the 1972 tests.

The 1974 tests with CGA 13586 utilized plots of 4 trees each of 'Manzanillo', 'Ascolano', 'Sevillano', and 'Mission'. CGA 13586 at concn of 1750, 2000, and 2250 ppm were used in each plot. Sprays were applied to 'Manzanillo' trees at Ivanhoe, California on Oct. 1 (32°C max; 13° min) and to 'Ascolano' trees at Madera on Oct. 2 (15°C max; 10° min). A rain of approx 24 mm fell the night of Oct. 1 in both districts. 'Sevillano' trees at Corning were sprayed Oct. 7 (30°C maximum; 15° minimum) and 'Mission' trees at Orland were sprayed Oct. 23 (24°C max; 13° min).

Tree shaking was done in each plot 8 days after the spray application using an Orchard Machinery Co. Shock Wave Shaker, fitted with a double expanded head, 14–15 star shaking pattern, and using 5 W4-LAB weights. Fruit removal percentage was calculated from the weight of fruit removed by the shaker and the amount of fruit left on the tree. Percentage leaf removal during shaking was obtained by using 20 tagged shoots around each tree on which leaf counts before spraying and after shaking were made.

Further tests were conducted to determine when fruit removal force reached a minimum after spraying. A series of 140 fruits in similar locations on each test tree was tagged. Sprays of CGA 13586 at 2500 ppm + 0.3% Regulaid, cycloheximide at 60 ppm + 0.3% Regulaid, and ethephon (Ethrel) at 2500 ppm + 0.3% Regulaid were then applied to separate branches. Fruits were also tagged on untreated branches. Twenty fruits were pulled and the pull force recorded every other day after spraying.

A series of tests was conducted in which different individual parts of fruiting shoots were treated separately with CGA 13586 at 3000 ppm and the fruit removal force was measured 8 days later. Parts treated were leaves only, fruit only, fruit peduncle only, and the entire fruiting shoot. This was done by

Table 1. Effect of CGA 13586 on fruit removal force and leaf drop on 'Manzanillo' olives. Sprays applied Oct. 24, 1972.

Concn of CGA 13586 (ppm)	Fruit removal force (g)	Leaf drop (%) Oct. 6	Leaf drop (%) Nov. 12
0	453 b[z]	0	1
500	272 ab	3	3
750	277 ab	2	5
1000	197 a	1	6
1500	141 a	2	9

[z]Mean separation by Duncan's multiple range test, 1% level.

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Table 2. Effect of CGA 13586 on percentage of fruits and leaves removed by shaking and on fruit removal force on 3 olive cultivars. Sprays applied Oct. 30, 1973; measurements taken on Nov. 8.

Olive cultivar	Variable	CGA 13586 concn (ppm) 0	1500	2000	2500	3000
Manzanillo	FRF (g)	663a[z]	457a	333a	213b	161b
	Fruit removal (%)	77a	76a	98b	98b	100b
	Leaf removal (%)	2a	2a	7ab	14b	7ab
Mission	Fruit removal (%)	51a	89b	96b	94b	92b
	Leaf removal (%)	2a	12a	4a	9a	7a
Sevillano	Fruit removal (%)	80a	85ab	90ab	96b	–
	Leaf removal (%)	6a	5a	13ab	20b	–

[z]Mean separation in rows by Duncan's multiple range test, 5% level.

dipping them in vials of solution except for the peduncle, which was painted with the solution.

A comparison of ethylene evolution from 'Manzanillo' fruits and leaves treated with 2500 ppm CGA 13586, with 2500 ppm ethephon, or with 60 ppm cycloheximide was made over an 11-day period. Branch units were sprayed with or dipped into these materials. Fruit and leaf collections were made at daily intervals for the first 5 days, with additional collections on the 7th and 11th days. Fruits and leaves were placed separately into 1 liter containers aerated at a flow of about 10 ml per min for ethylene determination. Ethylene concn was measured after about 4 hr using an Aerograph series 1200 gas chromatograph with a flame ionization detector and fitted with a 0.3 × 152 cm column packed with 60/80 mesh alumina.

A study was made of the pH effect on ethylene evolution from CGA 13586 solutions. Twenty μl (12.8 μmoles) of CGA 13586 was placed in Erlenmyer flasks with 5 ml of the appropriate buffer solution before sealing. Gas was withdrawn with a syringe and injected into the gas chromatograph as described above for ethylene determinations.

Small container-grown 'Manzanillo' trees in the greenhouse were sprayed with 3000 ppm solutions of CGA 13586 at different pH values from 5.0 to 8.5 to determine the abscission effects of CGA 13586 buffered to various pH levels. Buffers at both 3 mM and 50 mM concn were used. Trees were shaken vigorously by hand and the percentage leaf drop determined after a 7 day period.

Results

A significant difference from the control in fruit removal force was obtained by the use of CGA 13586 on 'Manzanillo' olives in 1972 but differences among concn were not significant (Table 1). Leaf abscission was minor.

Table 3. Fruit and leaf removal following spraying with CGA 13586 and shaking with a commercial tree shaker in orchard tests using 4 olive cultivars, 1974.

Cultivar	District	CGA 13586 concn (ppm) 0	1750	2000	2250
			Fruit removal (%)		
Manzanillo	Ivanhoe	71 a[z]	77 b	84 b	87 b
Ascolano	Madera	50 a	91 b	85 b	82 b
Sevillano	Corning	83 a	96 b	92 b	96 b
Mission	Orland	57 a	–	84 c	–
			Leaf removal (%)		
Manzanillo	Ivanhoe	2 a	10 b	21 c	15 b
Ascolano	Madera	1 a	43 b	44 b	40 b
Sevillano	Corning	16 a	22 a	23 a	25 a
Mission	Orland	6 a	–	17 b	–

[z]Mean separation in rows by Duncan's multiple range test, 5% level.

Table 4. Effect of CGA 13586 (3000 ppm) contact location on 'Manzanillo' olive fruit loosening after 8 days, 1974.

Treatment applied to:	Fruit removal Force (g)	% of control
Leaves only	389 a[z]	86
Fruit only (not cavity)	374 a	90
Fruit cavity (ca. 1 drop)	171 b	41
Fruit stem only	385 a	93
Whole branch	150 b	36
Control, no treatment	414 a	100

[z]Mean separation by Duncan's multiple range test, 5% level.

The effect of 4 concn of CGA 13586 on percentage fruit and leaf removal of four olive cultivars in 1973 trials is shown in Table 2, as is the effect on fruit removal force for 'Manzanillo'. Generally, 2000 to 2500 ppm was required for adequate fruit removal. Leaf removal was also generally minimal in these trials.

A significant difference in fruit removal was obtained in each case from the unsprayed trees in 1974 but differences among the three CGA 13586 concn were not significant (Table 3). Leaf removal comparisons between sprayed and unsprayed trees were also significant in each case except 'Sevillano'. The excessively high leaf removal for 'Ascolano' could possibly be explained by rains that occurred the night before the sprays were applied causing high humidity and subsequent high ethylene release following spraying.

Reduction in fruit removal force occurred most rapidly with CGA 13586, followed by ethephon, then cycloheximide, which did not reach the levels attained by the first 2 materials (Fig. 1). The effect from cycloheximide was delayed from 4 to 6 days after spraying, then progressed more slowly than the others. Minimum pull force values occurred 8 to 10 days after spraying.

It is important to initially tag the location of each fruit to be pulled in obtaining data of this nature. Some tagged fruits drop during the course of the test and can be given a zero pull force value. The dropped fruits will be missed if untagged fruits are randomly pulled and only the remaining attached fruits can be pulled toward the end of the test. Some of these apparently are missed or are unaffected by the spray and have high pull force values. Mean readings toward the end of the test will then show an increase, indicating that there is a tightening of the fruits to the stem.

Table 4 shows the reduction in fruit removal force when different portions of the fruiting shoot were treated with CGA

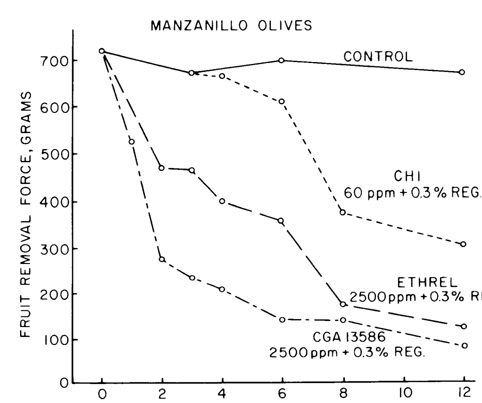

Fig. 1. Reduction in removal force of Manzanillo olive fruits following spray applications of three ethylene producing chemicals, CGA 13586, Ethrel (ethephon), and cycloheximide.

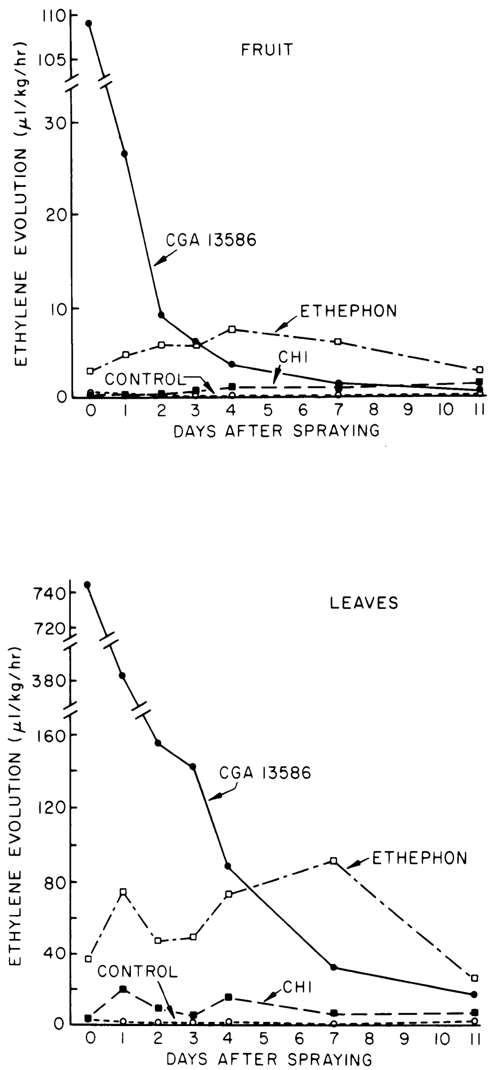

Fig. 2. Pattern of ethylene evolution from olive fruits (top) and leaves (bottom) treated with CGA 13586, ethephon, and cycloheximide.

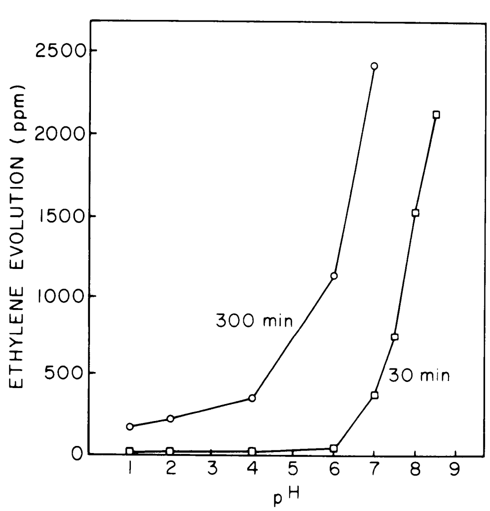

Fig. 3. Ethylene evolution from aqueous solution of CGA 13586 as affected by pH (after 30 min and after 300 min).

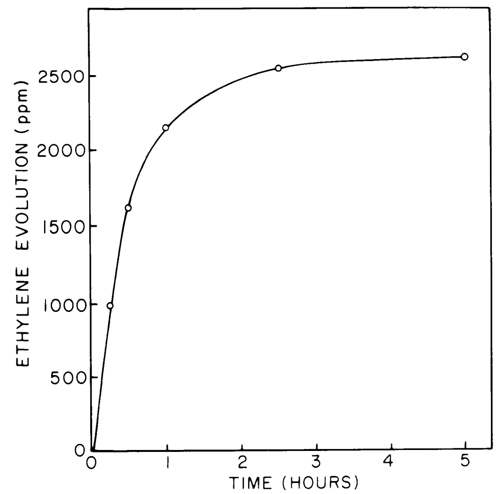

Fig. 4. Time course of ethylene evolution from aqueous solution of CGA 13586 buffered at pH 8.

13586. Significant indication of abscission induction occurred only when either the entire shoot was treated or when the solution was placed in the fruit cavity. This indicates that the action of the material lies in an ethylene influence directly on cells at the abscission layer and that a translocation effect is not involved.

Both leaves and fruits treated with CGA 13586 released ethylene at a considerably higher rate initially than those treated with ethephon or cycloheximide but the ethylene output from CGA 13586 treatments decreased to less than that from ethephon after 4 or 5 days (Fig. 2). Ethylene release after cycloheximide treatment remained relatively low throughout the measurement period.

Ethylene evolution from CGA 13586 increased very rapidly in aqueous solution with an increase in pH although the reaction proceeded slowly even at pH 1 (Fig. 3). The time course of ethylene evolved at pH 8 shown in Fig. 4 indicates that CGA 13586 had a half-life of only 20 min under these test conditions.

Influence of the pH of the CGA 13586 solution on olive leaf abscission is shown in Fig. 5. There was no significant difference from pH 5.5 to pH 8.5 with 3 mM buffers, the abscis-

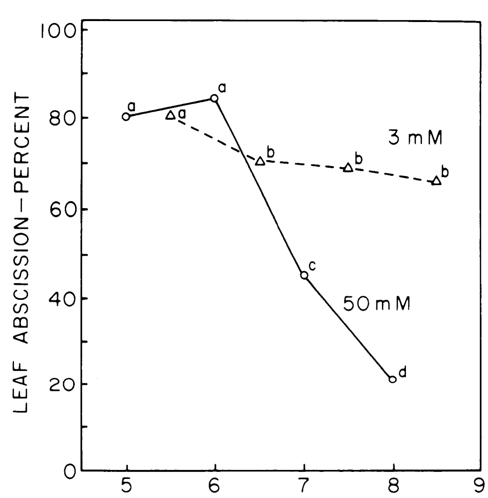

Fig. 5. Influence of pH of aqueous solution of CGA 13586 (3000 ppm) on 'Manzanillo' olive leaf abscission with buffers at 3 mM and at 50 mM concentrations. (Mean separation among pH values by Duncan's multiple range test, 5% level.)

sion percentage ranging from 70 to 80%. However, the highest leaf abscission (80 to 82%) occurred at pH 5 and 6 when 50 mM buffers were used with the abscission percentage decreasing considerably as pH increased, reaching a minimum at pH 8. It appears that ethylene is lost too rapidly in solutions above pH 6 to effectively cause abscission.

Discussion

The property of CGA 13586 as a releaser of ethylene results from its great reactivity with water, especially at high pH levels. The initial rate of ethylene evolution is much higher than that from ethephon. The rate of evolution per mole at a pH near 6 was found to be about 50 × as great as the rate from ethephon, the latter based on data of Warner and Leopold (5); the ratio is still higher at pH values above or below 6.

Hydroxyl ions are consumed and chloride ions are released when CGA 13586 reacts with water (4). This leads to acidification of the solution as the reaction proceeds and a consequent decrease in the rate of the reaction. Any excess HCl remaining during evaporation of a spray solution may be expected to volatilize and be lost along with the water.

CGA 13586 induces greater fruit abscission in olives but lower leaf abscission than ethephon or cycloheximide (2). The causes of this difference, as studied by ethylene evolution from fruits and leaves is not clear (Fig. 2). Initial rates of ethylene release from both fruits and leaves sprayed with CGA 13586 are much higher than those from fruits or leaves sprayed with ethephon but the ethylene output from plant parts treated with CGA 13586 decreased rapidly and is lower than from those treated with ethephon after 3 to 5 days. It is possible the prolonged presence of ethylene following ethephon sprays is more effective in causing leaf abscission than the shorter duration of high ethylene resulting from CGA 13586 with a much lower rate of ethylene production (2). Ethylene produced inside the tissue (as is very probably the case for the injury-stimulated ethylene from CHI) is probably effective in promoting abscission. Ethylene-releasing substances, such as CGA 13586 and ethephon, on the other hand, may release a large proportion of the ethylene at the tissue surface where much of it is lost to the atmosphere.

CGA 13586 shows promise as a suitable material for use in mechanical harvesting procedures with California table olive cultivars, since the ethylene released causes a marked reduction in fruit removal force and significant increases in fruit removal percentages following tree shaking. Some leaf removal occurred in the tests reported here but it was not excessive.

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Tissue Culture-induced Variation in Scented *Pelargonium* spp.[1]

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Abstract. ***In vitro*** **techniques were developed to regenerate plantlets (calliclones) from callus of scented geraniums (*Pelargonium* spp.). Calliclones were compared to plants derived from stem, root, and petiole cuttings of 5 cultivars. Plants from stem cuttings of all cultivars were uniform and identical to the parental clone. Plants from root and petiole cuttings were more variable with the amount of variation dependent upon cultivar. High variability was associated with calliclones. Aberrant types included changes in plant and organ size, leaf and flower morphology, essential oil constituents, fasciation, pubesence, and anthocyanin pigmentation. Calliclone variation was dependent upon clone and age of callus. Variability in calliclones was due to segregation of chimeral tissue, euploid changes, and heritable changes which may involve individual chromosomal aberrations or simple gene mutations. Variability of calliclones might be exploited for improvement of vegetatively propagated crops especially highly polyploid, sterile lines.**

Crop improvement is based on the creation of variable populations from which superior genotypes are isolated. Traditional plant breeding is dependent upon the sexual cycle, i.e., variation in the population is released and stored through the processes of meiosis and fertilization. Experience has shown that crop improvement utilizing this system may be remarkably efficient at first, but genetic advances become increasingly difficult with time. Certain unique combinations seem difficult to improve.

Dependence on the sexual system may impose a restraint to genetic improvement, especially severe for polyploid, vegetatively-propagated crops where the economic portion is not the seed. Crops cultured for their vegetative organs might be rendered more efficient by the complete elimination of the sexual apparatus, mainly as a conservation of biological function and energy. Further, the sexual system is difficult to control. Mild changes cannot be obtained except by backcross procedures which are time-consuming and unsuitable to many genetic systems. Changes obtained by mutagenic agents are random and the resulting plants are often chimeral. Desirable changes accompanied by a defective sexual system usually become lost or unavailable. Thus, genetic improvement is not directly available for completely sterile plants; the only variations that occur in these types are due to selection within spontaneous mutations (sports). Further, in polyploids, genetic gain is slow and

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