

THE INFLUENCE OF CARBON DIOXIDE AND ETHYLENE PREPLANT SEED TREATMENT ON RESPIRATION AND GERMINATION OF TOMATO AT LOW TEMPERATURES

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ABSTRACT

THE INFLUENCE OF CARBON DIOXIDE AND ETHYLENE PREPLANT SEED TREATMENT ON RESPIRATION AND GERMINATION OF TOMATO AT LOW TEMPERATURES

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The interaction of carbon dioxide (CO_2) and ethylene (C_2H_4) gas on imbibed tomato (Lycopersicon esculentum Mill.) seed as a preplant treatment was investigated. Imbibed tomato seeds (cv. Heinz 1439) were placed into sealed jars and treated with three levels of CO_2 and four levels of C_2H_4 . After 24 hours at 20°C the seeds were removed, air dried, and assayed for germination at suboptimal temperatures.

Monitored levels of gas during the treatment indicated that absorbing C_2H_4 with potassium permanganate resulted in reduced respiration as measured by CO_2 evolution when compared to treatments where C_2H_4 was present. The level of evolved CO_2 reached 0.3% after 24 hours and approximately 12% after six days.

Seeds treated with CO_2 and C_2H_4 and then dried have a carry-over effect which can be manifested later during germination at low temperatures.

There was little effect of treatments on speed or percent germination at temperatures above 15.5° C. Below this temperature the levels of CO₂ and C₂H₄ during the treatment became important. The most rapid germination at 15.5° C, 15-hour days and 10°C, 9-hour nights in soil and at 12.2°C continuous temperature in Petri dishes occurred when CO_2 was allowed to evolve during the initial treatment or when 5% CO_2 was complimented with additional (60 ppm) C_2H_4 . If CO_2 was absorbed by lime, the high treatment rate (30 and 60 ppm) of C_2H_4 was inhibitory to germination in both the soil and Petri dish assay.

It appears that a balance of the two gases must exist and that a combination of these can be successfully used before planting to increase the germination rate at low temperatures.

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INTRODUCTION

Direct seeding of vegetables has increased greatly over the past several years. Contributing to this development are: 1) increased cost of labor for transplanting, 2) the development of better planting units that can place small seeds accurately, 3) the development of good herbicides that reduce weed competition, 4) the decreased spread of plant bed diseases, 5) the trend toward larger farms and more mechanization.

Direct seeding of tomatoes (<u>Lycopersicon esculentum</u> Mill.) in particular has increased because of these developments. One of the main problems in direct seeded tomatoes has been the failure to obtain a desired plant population and distribution because of cold, wet conditions that often occur during the spring in temperate areas of the United States. Therefore, uniform germination and rapid emergence is needed in direct seeded tomatoes during these periods of unfavorable conditions for optimum production.

A great deal of work has been done with seed treatments, but most of this has been limited to overcoming dormancy in crops such as the legumes and lettuce. Less has been done to decrease the minimum germination temperature of nondormant seeds. Kotowski in 1926 (21) reported that the

percentage and speed of germination were not increased by using chemicals over distilled water. He used germinating temperatures of 25°C for parsnip and pepper and 11°C for spinach. These temperatures should not be limiting germination in these crops. Ells (11) reported that K₃PO₄ and KNO₃ treatment stimulated germination in tomatoes when seeds were exposed to 10°C night temperature. Over and Koehler (26) also discussed a method of treating tomato seeds with aerated nutrient solutions to hasten germination. They found that the effect depended partly on maintaining an osmotic concentration high enough to prevent germination during treatment and then drying the seed quickly before germination could occur.

Mayer and Poljakoff-Mayber report (24) on the use of KNO_3 , thiourea, Gibberellin and CO_2 to increase germination of seeds. In most cases these treatments were used to overcome dormancy or substitute for light or some other requirement.

Pollock and Toole in 1966 (29) found that lima beans are extremely sensitive to low-temperature injury during the early stage of imbibition. In 1969 Pollock (28) found that temperature sensitivity was controlled by the amount of water in the seed at the time that imbibition of water began. Increasing initial seed moisture to 20% eliminated temperature sensitivity. Christiansen (8) pregerminated cotton seeds at 31°C, redried them, and found that protection against chilling injury was retained. He later found that initial

seed moisture was the controlling factor and that seeds were sensitive to temperature only when imbibition was started at moisture levels below 14% (9). Phillips and Youngman (27) found that under a mean soil temperature below 20°C more seeds emerged as moisture content was increased from 8 to 11 or 14% in grain sorghum. Bleak and Keller (4) reported that germination, seedling emergence, and initial root and shoot elongation were all hastened by preplanting treatment applied to the seeds of seven forage species. Drosdov and Sokolova (10) found that treating flax seeds with boric acid solution before planting increased drought tolerance. Mart'yanova <u>et al</u>.(23) increased the fruit yield of tomatoes by soaking seeds in water for 30 hours and then drying before planting.

Carbon dioxide (CO_2) has been reported to overcome seed dormancy in a number of cases. Thornton in 1935 (33) showed that 40% CO_2 can break thermodormancy in lettuce seed that had been induced by 35°C treatment. Carr in 1961 (6) listed a number of other cases where CO_2 had broken dormancy in seed. Usually, the level of CO_2 needed to break dormancy is quite high, and many workers feel that these concentrations are too high to be encountered by seeds in nature. However, promotive effects of lower CO_2 concentrations have been reported. Hart and Berrie (15) found that 3% CO_2 overcame the inhibition of light in <u>Avena fatua</u> L. Ballard in 1958 (3) reported that .5 to 5% CO_2 can overcome dormancy in subterranean clover seed. He suggests that because of the capacity of imbibed

seed to produce CO₂, the number of seeds in relation to the volume of air around them is important. Under field conditions, mechanical impedence of the soil is frequently overcome much more effectively by an aggregate of germinating seeds than by single seedlings (20).

Esashi and Leopold (12) and Ketering and Morgan (18) have shown that imbibed seeds of subterranean clover and peanuts produce $C_{2}H_{4}$ as well as CO_{2} and that their germination is promoted by both of these substances, probably in an independent manner. In comparing three varieties (two dormant and one non-dormant) of clover, Esashi and Leopold found that more $C_{2}H_{4}$ is produced by the non-dormant variety and that the production of $C_{2}H_{4}$ markedly preceeds the first emergence of the radicle.

Abeles and Lonski (2) reported that non-dormant lettuce seeds produced ten times as much C_2H_4 as dormant ones and concluded that this is an effect of germination rather than a controlling factor. They also reported that CO_2 did not act as a competitive inhibitor of C_2H_4 in lettuce seed germination as it does in a number of other processes such as root (7) and stem (5) growth inhibition, fruit ripening (19), celery blanching (22), flower wilting (30), abscission (1), hook opening (17), and peroxidase formation (13).

Negm, Smith and Kumamoto (25) reported on the interaction of CO_2 and C_2H_4 in completely overcoming thermodormancy of lettuce seeds at 35°C. The combination is effective if it is added to seeds either at the start or after several days

of imbibition. The action of C_2H_4 is dependent upon the CO_2 level present in the atmosphere surrounding the seeds. When CO_2 is trapped by KOH the C_2H_4 effect is essentially nil. Takayanagi and Harrington (32) found that C_2H_4 gas around the seed will restore rape seeds that are declining in vigor to almost original vigor. They suggest that degradation of the C_2H_4 producing system or insufficiency of the substrate(s) which normally give rise to C_2H_4 may occur in aged seeds. Harrington (14) speculated on the possibility of treating seeds with (2-chloroethyl) phosphonic acid (ethephon) to provide a C_2H_4 atmosphere around the germinating seed, thereby improving vigor and speeding emergence.

Stewart and Freebairn (31) state that it is possible C_2H_4 may be an intermediate between auxin and the other physiological responses attributed to auxin in some plants. Jones (16) concluded that C_2H_4 does not affect *a*-amylase synthesis in barley but rather that its effect is on *a*-amylase secretion from aleurone cells.

Varner <u>et al</u> (34,35) have shown that α -amylase is produced <u>de novo</u> in aleurone layers following treatment with gibberellic acid (GA₃). Villiers (36) states that dormancy may be due to the presence of growth inhibitors, the absence of growth promoters or a combination of both. The growth promoters referred to most often are the gibberellins.

In an attempt to find a treatment that would stimulate germination of tomatoes at suboptimal temperatures, the following experiments were performed during the period 1 April

to 1 September, 1972. Section I deals with the interaction of CO_2 and C_2H_4 on tomato seeds, Section II deals with the effect of ethephon on tomato seed germination and Section III deals with the effect of GA_3 on cabbage and tomato seed germination.

MATERIALS AND METHODS

Section I

CO2 and C2H4 Preplant Treatments

Tomato seeds (cv. Heinz 1439) were treated with various concentrations of CO_2 and C_2H_4 gas in closed containers. A piece of 7-cm diameter Whatman #1 filter paper was placed in the bottom of a .473-L Mason jar and was soaked with 3 ml distilled water. Approximately 500 dry tomato seeds (measured by a volume of 2.3 cc) were placed on the moist paper in each jar. A lid with tightly fitted serum caps was used to seal each jar (Fig. 1). The serum caps were used so that CO_2 and C_2H_4 could be added with a syringe and samples of gas could be taken periodically. Twelve jars were used for three levels of CO_2 and four levels of C_2H_4 . An additional treatment consisted of imbibing seeds with distilled water for this period in an unsealed jar. The treatments were then held in a room at 20°C for 24 hours.

The three levels of CO₂ were obtained by 1) absorbing CO₂ with lime. The amount, 1.25 grams, of lime was placed on a 4.11-cm square plastic weighing tray over the seed to absorb the CO₂ from around the seed; 2) allowing the CO₂ to evolve naturally; 3) adding CO₂ with a syringe to reach a level of 5%. To do this, 26 cc of air was withdrawn from the jar and the same volume of 100% CO₂ was added. The physical

Figure 1. Jar used to treat imbibed tomato seed

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characteristics of gases are dependent on pressure and temperature; therefore, through several trials, it was determined that 26 cc of 100% CO₂ was needed to obtain this level.

The four levels of C_{2H4} were obtained by 1) absorbing C_{2H4} with potassium permanganate. About 1.25 grams of potassium permanganate coated on perlite granules* was placed on weighing trays to absorb the C_{2H4} from around the seed; 2) allowing the C_{2H4} to evolve; 3) adding C_{2H4} to obtain approximately 30 ppm and 4) adding C_{2H4} to obtain 60 ppm. The C_{2H4} was added by diluting 1 cc of 100% C_{2H4} in a 30-cc syringe and then taking 0.5 cc from this and injecting it into a jar to obtain 30 ppm. One cc was used to obtain 60 ppm.

The levels of both gases were monitored at the beginning of the experiment and at 6, 12, and 24 hours. Samples of the gas were withdrawn from each jar with a 1-ml plastic syringe inserted through the serum cap and assayed using gas chromatography. The CO_2 concentration was determined on a Perkin-Elmer 154B gas chromatograph employing a column of silica gel. C_2H_4 analysis was performed employing a Varian 1720 gas chromatograph equipped with a .32 x 76.2-cm column of activated aluminum connected to a flame ionization detector. The results of the gas chromatography are shown in Table 1.

After treatment for 24 hours at 20°C, the seeds were removed from the jars and placed in open Petri dishes to dry

* Trade name, "Purafil." H. E. Burroughs and Assoc., Inc., Chamblee, Georgia.

for 24 hours at room temperature. Germination tests of the seeds were run by placing them in Petri dishes and cups. Fifty seeds were counted and placed in a Petri dish on 9-cm diameter Whatman #2 filter paper which had been soaked with 3 ml distilled water. These were then placed in a dark growth chamber with a continuous temperature of 12.2°C. Twenty-five seeds were counted and placed in a 236-cc styrofoam cup which contained 176 cc sterilized greenhouse soil which had been moistened with 30 ml distilled water. The seeds were covered with 30 cc of soil. The cups were then placed in a growth chamber with 15 hours of light at 15.5°C and 9 hours dark at 10°C. To minimize the effect of unequal chamber temperatures, the cups were rotated daily. Watering was done on a daily basis as needed. As a control, untreated unimbibed seeds were also planted in both tests. Each treatment was replicated five times.

Germination counts in the Petri dishes were made at three-day intervals and germination consisted of visible radical emergence. In the cups, counts were made daily as the seedlings emerged from the soil.

An analysis of variance was performed on each experiment, and treatment means were compared using Duncan's Multiple Range Test. A factorial AOV was used in comparing the first 12 treatments and a randomized complete block design was used in comparing the water-imbibed and untreated seeds as additional treatments.

In an additional experiment, the treated tomato seeds

were kept in the sealed jars for six days and the amount of CO_2 and C_2H_4 was monitored regularly to determine the levels of these gases over an extended period.

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MATERIALS AND METHODS

Section II

Ethephon treatment

Tomato seeds (cv. Heinz 1439) were soaked in solutions of 0, 40, 80 or 160 ppm (2-chloroethyl) phosphonic acid (ethephon) for 30 or 60 minutes. The seeds were then removed and dried at room temperature for 24 hours. The dried seeds were then counted with 100 seeds being placed in each Petri dish, 50 seeds in each cup and 50 seeds planted in each plot in the field. In each case there were five replications.

The Petri dishes were prepared the same as in Section I. In the cups, Jiffy Mix was used instead of soil. These were then placed in a dark growth chamber at 12.5°C. The field plots were planted with a hand-operated Planet Jr. seeder adapted with a cone distributor. Seeding was done on May 10 and the soil temperature was 16.6°C at the 5-cm level. The soil was a Fox sandy loam. Germination counts were made periodically in the Petri dishes and cups and, after 19 days, in the field.

Granular ethephon (5% G) at the rates of 0, 0.28, 11.2, and 44.8 kg/ha were applied as an in-row seed treatment on two cultivars of tomato (Heinz 1350 and MH 1) and one cultivar of cabbage (Golden Acre). These were seeded on a sandy

loam soil at Sodus, Michigan on July 5. Germination was determined July 13.

Granular ethephon was also mixed with dry Jiffy Mix to dilute the chemical and apply it as a seed treatment for tomatoes (cv. Heinz 1439) in cups. The rates used were 0, 1.12, 2.24, 4.48, 8.96 and 17.92 kg/ha. These were then placed in growth chambers with 15.5°C for 15-hour days and 10°C for 9-hour nights. The seeds were planted July 14 and germination counts were made July 24 and 27.

MATERIALS AND METHODS

Section III

Gibberellic Acid (GA₃)

Tomato (cv. Heinz 1439) and cabbage (cv. Golden Acre) were treated with various concentrations of gibberellic acid (GA₃) for one hour. The concentrations used were 0, 0.5, 1, 2, 4 and 8 x 10^{-4} M.

After treatment, the seeds were removed and air dried for 24 hours. Seeds were then counted and placed in styrofoam cups in Jiffy Mix as in Sections I and II. The cups were then placed in the growth chamber at 15.5°C for 15-hour days and 10°C for 9-hour nights. Germination counts were made on cabbage 10 days after seeding and on tomatoes 14 days after seeding.

RESULTS AND DISCUSSION

Section I

CO2 and C2H4 Preplant Treatments

The monitored levels shown in Table 1 indicate that there was very little if any change in C_2H_4 level during the 24-hour treatment. However, where potassium permanganate was used to absorb the $C_{2}H_{4}$, the CO_{2} evolution is reduced about 50% compared to the other treatments containing evolved or added C_{2H_4} after 24 hours. The results given in Figure 2 are averages of five repeated treatments and demonstrate that C_2H4 has an effect on respiration in imbibed tomato seeds as measured by CO₂ evolution. To verify that this was due to a difference in respiration and not CO₂ absorption by the potassium permanganate, several jars containing a known concentration of CO2 were set up with and without this absorber. Samples were taken periodically and no differences were found over 72 hours (see Table 2).

The levels of CO_2 over the six-day period are given in Figure 3. The treatment containing potassium permanganate shows a lower CO_2 evolution up to 96 hours. However, after 96 hours the rate increased and after 144 hours the CO_2 level was the same as the treatment which contained 30 ppm C_2H_4 .

Table 1.	Levels of carbon	dioxid	e and et	thylene 1	monitore	ed during	tomato	seed trea	tment
Tre	atment		CO	%			C2H4	ppm	
C02	C2H4	0 hrs	6 hrs	12 hrs	24 hrs	0 hrs	6 hrs	12 hrs	24 hrs
absorbed	absorbed	0	0	0	ο	0	0	0	ο
absorbed	evolved	0	0	0	0	0	С	0	0
absorbed	30 ppm	0	0	0	0	37.39	40.59	38.18	39.63
absorbed	60 ppm	0	0	0	0	57.47	61.55	61.47	60.81
evolved	absorbed	0.07	0.05	0.04	0.15	0.27	0.09	0.05	0.02
evolved	evolved	0.04	0.06	0.10	0.30	0	0.01	0.01	0.03
evolved	30 ppm	0.05	0.05	0.09	0.29	41.71	37.32	36.56	36.89
evolved	60 ppm	0.02	0.05	0.09	0.30	58.09	61.55	58.88	60.81
5%	absorbed	4.65	4.32	4.22	4.22	0	0	0	0.03
5%	evolved	5.78	5.62	5.58	5.77	0.01	0.01	0	0.03
5%	30 ppm	5.91	5.75	5.81	5.94	31.52	34.05	33.0	32.80
5%	60 ppm	5.64	5.53	5.84	5.64	57.16	63.51	60.82	60.12

Table 2.	Monitored lev after 72 hour potassium per	vels of % CO ₂ s in Mason ja manganate	at the begin ars with and	ning and without
Treatment	0 hc	ours 72	hours	Decrease
Without KM	In04 J	95	1.58	0.37
With KMn0 ₄	. 1		1.39	0.46

Figure 2. CO₂ measured over 24 hours during seed treatment



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Figure 3. CO₂ measured over a six-day period during seed treatment



At this time the oxygen level had decreased and could have been limiting.

These same trends are noticed in the treatments containing 5% CO₂ at the beginning. The treatment which contained 5% CO₂ and the C₂H₄ absorber lagged behind in respiration for 72 hours but then increased and resulted in slightly more CO₂ evolved than the treatment containing added CO₂ and 30 ppm C₂H₄. In both cases, the greatest CO₂ evolution occurred if C₂H₄ had been allowed to evolve freely. In this extended treatment, the evolved C₂H₄ reached a level of 0.2 ppm after six days at all levels of CO₂.

Germination results

All of the gas and water-imbibed treatments were better than the control at low germination temperatures in the Petri dish study. The results in Table 3 indicate that the best germination occurred where C_2H_4 was absorbed and the CO_2 allowed to evolve or if 60 ppm C_2H_4 and 5% CO_2 were added. The slowest germination occurred in the untreated control, the water-imbibed seeds and in the treatments where CO_2 was absorbed with lime and 30 and 60 ppm C_2H_4 added.

Table 4 is a list of the results of the germination counts in the soil emergence test. The evolved CO_2 and the added (5% CO_2 and 60 ppm C_2H_4) gas treatments showed the best early germination. The slowest germination was in the control and where CO_2 was absorbed and 30 or 60 ppm C_2H_4 was added after 13 days in Petri dishes and eight days in soil.

The high concentration of C_2H_4 in the absence of CO_2

Table 3. Effect of carbon dioxide and ethylene on number of germinated tomato seeds at 12.2°C in Petri dishes (50 seeds total)

Treat	ment	Days after seeding		
co ₂	C ₂ H ₄	13	16	19
absorbed	absorbed	2.2 bc	9.8 cdef	19.6 bc
absorbed	evolved	4.0abc	17.8abc	27.0ab
absorbed	30 ppm	1.2 c	12.2 bcde	22.0abc
absorbed	60 ppm	1.6 c	10.2 bcdef	18.2 bc
evolved	absorbed	5.0ab	21.6a	30.0a
evolved	evolved	3.6abc	15.8abcd	25.8ab
evolved	30 ppm	1.8 bc	10.0 cdef	21.4abc
evolved	60 ppm	3.0	16.8abcd	26.4ab
5 %	absorbed	2.2 bc	9.0 def	19.0 bc
5%	evolved	4.2abc	15.6a bcd	27.0 a b
5%	30 ppm	2.0 bc	6.0 ef	14.6 c
5%	60 ppm	5.6a	18.4 ab	29.2a
imbibed		1.6 c	8.8 def	15.6 c
control		1.0 c	2.6 f	5.4

Means within columns followed by common letters are not significantly different at the 1% level (Duncan's Multiple Range Test).

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Table 4. Effect of carbon dioxide and ethylene on the number of germinated tomato seeds in soil at 10°C night and 15.5°C day temperatures (25 seeds total)

Treatment			Days aft	er plan	ting	
co ₂	C ₂ H ₄	8	9	10	11	15
absorbed	absorbed	1.8abc	4.4abcd	16.6a	18.4a	23.2a
absorbed	evolved	2.8abc	5.4abcd	14.2a	15.6a	21.6 b
absorbed	30 ppm	0.6 c	2.8abcd	15.0a	16.8a	23.2a
absorbed	60 ppm	1.0 bc	3.2abcd	14.2a	15.6a	22.8a
evolved	absorbed	3.6abc	7.0ab	14.8a	16.0a	21.8 b
evolved	evolved	4.6a	6.0abc	14.8a	16.8a	22.4 b
evolved	30 ppm	1.6abc	3.4abcd	15.0a	16.0a	22.0 b
evolved	60 ppm	4.0ab	7.2a	16.0a	17.6a	22.0 b
5 %	absorbed	2.6abc	5.0abcd	16.6a	17.0a	22.6ab
5%	evolved	1.4 bc	5.2abcd	15.2a	15.8a	21.0 b
5 %	30 ppm	1.0 bc	2.4 cd	15.6a	16.0a	22.2 b
5%	60 ppm	3.2abc	5.2abcd	14.0a	15.2a	21.4 b
imbibed		1.4 bc	2.6 bcd	14.2a	15.4 a	21.4 b
control		0.6 c	1.2 d	6.2	7.2	18.0 Ъ

Means within columns followed by common letters are not significantly different at the 1% level (Duncan's Multiple Range Test). is inhibitory to early germination in both of these tests. In the soil, these treatments recovered and after 15 days, there was no difference in the total number germinated.

The treatments where the 30 ppm of C_2H_4 was added tended to have slower germination than the corresponding evolved and higher concentration of C_2H_4 .

In analyzing the 12 gas treatments as a factorial experiment there was no significant interaction between the different levels of CO_2 and C_2H_4 in the soil germination test. Table 5 is a comparison of the mean effects of the three levels of CO_2 over all levels of C_2H_4 and the mean effects of the four levels of C_2H_4 over all levels of CO_2 on germination in soil. This indicates that at the first count there was faster germination where CO_2 accumulated and there was inhibition where C_2H_4 at 30 ppm was added. However, two days later there were no significant differences in number of emerged seedlings.

In the Petri dish study there were significant differences due to interaction of CO_2 and C_2H_4 treatments (Table 6). The same problem of inhibition occurred where the 30 ppm concentration of C_2H_4 was used, similar to that expressed in the soil emergence test. Treatments with evolved CO_2 and absorbed C_2H_4 or 60 ppm added C_2H_4 resulted in faster germination but after 19 days from seeding, there were smaller differences among all treatments.

C			
	Day	<u>s after seed</u>	ling
	8	9	10
Level of CO ₂			
absorbed evolved 5% added	1.55 a 3.45 b 2.05 a	3.95 b 5.90 a 4.45 s	15.00 a 15.15 a 15 35 a
Level of C _{2H4}	2.05 4	4143 u	13:33 4
absorbed evolved 30 ppm 60 ppm	2.67 a 2.93 a 1.07 b 2.73 a	5.47 a 5.53 a 2.87 b 5.20 a	16.00 a 14.73 a 15.20 a 14.73 a

Table 5. Effect of carbon dioxide and ethylene on germination of tomato seeds in soil at 10°C night and 15.5°C day temperatures (25 seeds total)

Means within columns followed by common letters in each subcolumn are not significantly different at the 5% level (Duncan's Multiple Range Test).

Table 6. Effect of carbon dioxide and ethylene on germination of tomato seeds in Petri dishes at 12.2°C (50 seeds total)

13 days	13 days after planting				C2H	ł4			
		abso	rbed	evo	lved	30	ppm	60	ppm
	absorbed	2.2	abc	4.0	abc	1.2	с	1.6	Ъс
co ₂	evolved 5%	5.0 2.2	ab abc	3.6 4.2	abc abc	1.8 2.0	bc abc	3.0 5.6	abc a
16 days	after planting				C ₂ I	Ŧ4			
		absor	rbed	evo	lved	30	ppm	60	ppm
	absorbed	9.8	bc	17.8	ab	12.2	bc	10.2	bc
co ₂	evolved 5%	21.6 9.0	a bc	15.8 15.6	ab ab	10.0 6.0	bc c	16.8 18.4	ab ab
19 days	after planting				C ₂ 1	14			
		abso	rbed	evo	lved	30	ppm	60	ppm
co ₂	absorbed evolved 5%	19.6 30.0	abc a	27.0	ab ab	22.0 21.4	abc abc	18.2	bc ab
	ه/ د	13.0	auc	27.0	au	T# • 0	Ľ	47.L	au

Means followed by common letters at the same date are not significantly different at the 1% level (Duncan's Multiple Range Test).

RESULTS AND DISCUSSION

Section II

Ethephon Treatments

There were no major differences in seed germinated due to liquid ethephon treatments. The results given in Table 7 are averages of germinated tomato seeds taken on May 29.

The results of granular ethephon applications shown in Table 8 indicate that there was no stimulation at the low rates of application and that the high concentration was inhibitory to germination. It is possible that high soil temperature results in a greater release of C_2H_4 from ethephon which causes inhibition of germination or toxic effects in the seeds. In the growth chamber study there was no effect of the chemical treatment (Table 9).

Because of lack of apparent beneficial results, no further work was done with these approaches.

Ethephon rate (ppm)	Time of imbibition (min.)	Average number of seeds germinated
0	30	24
0	60	19
40	30	27
40	60	23
80	30	25
80	60	21
160	30	22
160	60	22
untreated contr	rol	18

Table 7. Effect of ethephon treatments on germination of tomato seeds in field plots (50 seeds total per plot)

There are no significant differences

	No.	emerged see	dlings $\frac{1}{}$
Ethephon rate kg/ha	Tomato (H 1350)	Tomato (MH 1)	Cabbage (Golden Acre)
0	63.2 ab	45.8 a	31.0 a
0.28	67.0 a	37.5 ab	29.0 a
2.8	46.2 b	27.2 ab	9.8 b
11.2	24.0 Ъ	12.0 bc	1.8 b
44.8	2.8 c	0.5 c	0.8 ъ

Table 8. Effect of granular ethephon on seed germination of two varieties of tomatoes and one of cabbage on a sandy loam soil.

 $\frac{1}{Counts}$ made eight days after application.

Means within columns followed by common letters are not significantly different at the 1% level (Duncan's Multiple Range Test).

temperatu	ires (50 seeds total)	seeded July 14
Ethephon rate kg/ha	No. germin July 24	ated seeds July 27
0	9.2	29.8
1.12	11.6	32.2
2.24	8.4	37.8
4.48	7.6	31.6
8.96	6.2	33.4
17.92	3.8	23.8

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Table 9. Effect of granular ethephon on germination of tomato seeds at 15.5°C day and 10°C night temperatures (50 seeds total) seeded July 14

There are no significant differences.

RESULTS AND DISCUSSION

Section III

Gibberellin Treatments

Gibberellic acid at the low concentration $(0.5 \times 10^{-4} \text{ M})$ gave the greatest stimulation for germination in cabbage and at the high concentration $(8 \times 10^{-4} \text{ M})$ stimulated tomato germination (Table 10). At the time of these experiments, these results did not seem as important or as significant as those observed in Section I. Therefore, no attempt was made to follow up on these observations. However, since there is promise of beneficial results with GA, there might be some interaction between this hormone and the effects of CO₂ and/or C2H4 as shown in Section I.

GA concentration	Percent germinated		
(M)	Tomatoes	Cabbage	
0.5×10^{-4}	68.5 Ъ	85.2 a	
1×10^{-4}	62.2 bc	75.6 ab	
2×10^{-4}	69.2 b	78.0 ab	
4×10^{-4}	66.0 bc	80.4 ab	
8×10^{-4}	93.0 a	80.6 ab	
imbibed control	56.6 c	82.0 ab	
control	59.4 c	71.6 b	

Table 10. Effects of GA₃ on seed germination of tomato and cabbage seed at 15.5°C day and 10°C night temperatures

Means within columns followed by common letters are not significantly different at the 1% level (Duncan's Multiple Range Test).

SUMMARY AND CONCLUSION

One of the most interesting results of these experiments is the effect of $C_{2}H_{4}$ on the rate of respiration as measured by the CO_{2} evolved. Carbon dioxide is a known competitive inhibitor of $C_{2}H_{4}$ action (1, 5, 7, 13, 17, 19, 22, 30); but in comparing treatments with absorbed $C_{2}H_{4}$ and evolved CO_{2} with other treatments, it appears that $C_{2}H_{4}$ has an effect on the production of CO_{2} in imbibed tomato seeds. Therefore, it seems reasonable to expect that some treatment with $C_{2}H_{4}$ can be used to increase respiration which, in turn, would release energy to be used in the germination process. This could be very important where low temperatures limit rapid and uniform seed germination.

Another interesting observation is that seeds treated with CO₂ and C₂H₄ for a period and then dried exhibit a carry-over effect which can be manifested later under certain environmental conditions.

As temperatures increased above 15.5° C, there was little effect of treatment on rate of germination; but below this temperature, the level of CO₂ and C₂H₄ became important. The most rapid germination occurred if the CO₂ was allowed to evolve freely and if the high rate of added CO₂ was complimented with additional C₂H₄. Therefore, it appears that a balance of the two gases must exist for optimum germination

. . at low temperatures.

Imbibed seeds have a great potential for the production of CO_2 . In the extended treatment for six days, the CO_2 level reached about 12%. Since CO_2 can stimulate germination, this could explain the lower germination in direct seeded crops where seed is spaced singly and the greater germination in seeds planted close together (3). This preplant treatment with CO_2 and C_2H_4 as described in Section I might be used to stimulate germination later when planted in the field due to the carry-over effect.

Other means of stimulating germination in seeds have been found such as the gibberellins (20, 24), nutrient salts (11, 26), etc. Since these work in a different way, the action of CO₂ and/or C₂H₄ could be used to compliment them and cause even greater stimulation under adverse conditions. Because some treatments have a carry-over effect on later germination, it may be possible to apply these methods, dry the seeds, and later plant them in the field. Whether or not this can be done on a commercial scale depends on the degree of stimulation that can be achieved and whether or not the expense can be recovered in final stand and yield. From the results presented here, it appears that these treatments show a great deal of promise and more work in this area would be valuable.

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APPENDIX

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LIST OF BINOMIALS OF PLANTS USED IN THE LITERATURE REVIEW

Barley	<u>Hordeum vulgare</u> L
Bean, lima	<u>Phaseolus</u> <u>lunatus</u> L
Celery	<u>Apium graveolens</u> L
Clover, subterranean	<u>Trifolium</u> <u>subterraneum</u> L
Cocklebur	<u>Xanthium</u> sp. L
Cotton	<u>Gossypium</u> sp. L
Flax	<u>Linum</u> sp. L
Lettuce	<u>Latuca sativa</u> L
Parsnip	<u>Pastinaca</u> <u>sativa</u> L
Peanut	<u>Arachis hypogaea</u> L
Pepper	<u>Capsicum</u> sp. L
Rape	<u>Brassica</u> <u>napus</u> L
Sorghum	Sorghum vulgare Pers.
Spinach	<u>Spinacia</u> <u>oleracea</u> L

