

AN ABSTRACT OF THE THESIS OF

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Title THE EFFECTS OF CIPC VAPORS ON DODDER (CUSCUTA  
SPP.) SEEDLINGS

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The use of CIPC (isopropyl N-(3-chlorophenyl) carbamate) to control dodder (Cuscuta spp.) in alfalfa is an established practice. A rate of six pounds per acre applied to moist soil has given good dodder control lasting from four to six weeks.

The purpose of this study was to determine the effect of CIPC vapors on dodder seedlings. In order to obtain a relatively high percent germination of dodder seeds, the seeds were scarified with concentrated sulfuric acid and planted in sterile soil. Since dodder is a parasitic plant it was necessary to find a suitable host plant. Alfalfa and carrots were used in initial experiments and toothpicks were substituted for the host plants in later experiments. It was found that dodder seedlings would wrap around toothpicks and haustoria would develop. Control of dodder seedlings was measured by counting the number of wrapped or attached dodder seedlings on a suitable host.

Dodder seedlings were exposed to CIPC vapors in a closed

plastic vapor trap system. Twenty percent granular CIPC at six pounds per acre was applied to moist soil within the vapor trap. Exposure of dodder seedlings to CIPC vapors for 2, 4, 8, 16, 32, and 64 hours indicated that at least 16 hours exposure was necessary to prevent seedlings from wrapping. A similar experiment was carried out using CIPC at six pounds per acre on dry soil. Complete control of dodder seedlings was obtained in each experiment.

Dodder seedlings were grown in 12 x 75 mm test tubes. They were then exposed to CIPC vapors released from CIPC granules in the open greenhouse. The test tubes provided assurance that all contact between CIPC and the dodder must have been as a vapor. In all cases the seedlings failed to wrap around the toothpicks.

Data from a field experiment using the test tube method and alfalfa as a host showed positive dodder control as a result of CIPC vapor toxicity.

An attempt was made to determine whether CIPC in the soil solution or CIPC vapors were most active in controlling dodder seedlings. In all cases seedlings were isolated from CIPC in soil solution. The vapors released did prevent wrapping of dodder seedlings on the toothpicks. A suitable method was not found which would assure only exposure of dodder seedlings to CIPC in the aqueous form.

The evidence collected shows that CIPC vapors are important in the control of dodder seedlings.

THE EFFECTS OF CIPC VAPORS ON  
DODDER (CUSCUTA SPP.) SEEDLINGS

by

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# THE EFFECTS OF CIPC VAPORS ON DODDER (CUSCUTA SPP.) SEEDLINGS

## INTRODUCTION

Dodder (Cuscuta spp.) is considered one of the most serious weed pests in alfalfa grown for seed. It is not uncommon for this weed species to reduce alfalfa seed production by 50% or more. Not only is it a troublesome pest to alfalfa seed growers but it is also known to transmit virus diseases.

The herbicide isopropyl N-(3-chlorophenyl) carbamate (CIPC) has been found to be effective in controlling this weed pest in alfalfa. Results of field tests with CIPC can be found quite extensively in the literature. A considerable amount of basic research has also been conducted on CIPC activity. However, there has been a major gap in the knowledge of CIPC volatility and its toxicity to dodder seedlings.

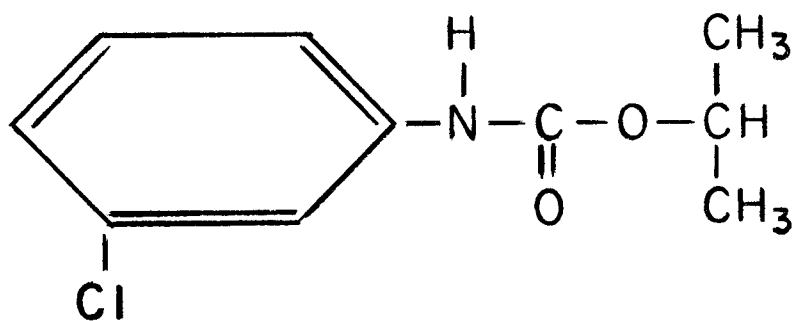
Lee and Timmons (35) reported that CIPC applied to a moist soil killed dodder seedlings selectively in alfalfa. Since this early work, other investigators have carried out extensive research on CIPC as it relates to dodder control. As with most research, several questions were brought up by their results. It was the intent of this research to extend the work of Dawson (11) on CIPC activity, especially volatility and its action on dodder seedlings. Other

objectives of this research program were: (a) to find a suitable substitute host for the dodder seedling thereby eliminating growing a host plant, (b) to observe the effects of CIPC vapors in a closed system, open greenhouse system, and under field conditions, (c) to develop a technique to expose the dodder seedling only to CIPC vapors, (d) to determine if CIPC vapors were indeed lethal to dodder seedlings.

## LITERATURE REVIEW

Physical and Chemical Properties

The structural formula for isopropyl N-(3-chlorophenyl) carbamate (CIPC) is given below:



CIPC

ISOPROPYL N-(3-CHLOROPHENYL)  
CARBAMATE

It is an ester that will slowly hydrolyze in acidic or basic conditions to yield m-chloro-aniline, carbon dioxide, and isopropyl alcohol (7).

CIPC has a molecular weight of 213.6 with a melting point of 38°C to 40°C. It is slightly soluble in water (108 ppm at 20°C) and is soluble in alcohols, hydrocarbons, chlorinated hydrocarbons, ketones, esters, and anhydrous ammonia (7).

### Soil Adsorption of CIPC

The activity and longevity of many herbicides are influenced by the adsorptive properties of the herbicide. CIPC is very tightly adsorbed to mineral and organic soil colloids (23, 24, 44). Danielson demonstrated the difference in the adsorptive capacities of various clay colloids. He found that there was little CIPC vapor loss from highly adsorptive carriers such as activated charcoal (8). Parochetti and Warren found that the adsorptive capacity of the soil is one of the major factors influencing volatility of IPC (isopropyl N-phenyl-carbamate) and CIPC (44). Clay and organic matter tend to bind the herbicide reducing loss through volatility. As the exchange capacity and the clay content or, organic matter content increases, vapor losses decrease from soils whether saturated or at field capacity (44). Vapor loss and soil diffusion are negligible in high organic and muck soils (15).

### Resistance to Leaching

CIPC does not move readily in the soil (43, 47, 49). Smith and Ennis reported that the movement of CIPC in soil varies with the formulation and that most of the CIPC is concentrated in the top inch (49). Ogle and Warren have demonstrated that CIPC does not leach when surface water is applied (43).

### Microbial Degradation

CIPC does not leach or move in the soil but its phytotoxic activity persists only for a short duration. Microbial degradation is one reason for this short period of activity (43). Kaufman and Kearney found that soil microorganisms degrade CIPC in perfused muck soil (27). The rate of degradation is a function of soil enrichment (27).

### Volatility

Vaporization or volatility of a compound is the evaporation from solid or liquid to gas at a given temperature.

Vapor loss of a herbicide can be important in the following ways:

1. The need to increase the rate of herbicide to get adequate weed control.
2. Crop injury from herbicide vapors.
3. Selective weed control if weeds are more sensitive to herbicide vapors than the crop plant.

Linder, Shaw, and Marth reported that the volatile carbamates, IPC and CIPC, required higher rates in order to obtain satisfactory weed control (37). The reason for the increased rates was to compensate for vapor loss. However, carbamates with lower vapor activity were less injurious to certain crop plants than the carbamates with



higher vapor activity such as IPC and CIPC (37).

Higher rates of EPTC (ethyl N, N-dipropylthiolcarbamate) are required when not incorporated into the soil due to loss of herbicidal activity by vaporization (1). It has been demonstrated both in the greenhouse and in the field that CIPC is volatile and that the vapors possess herbicidal activity (11, 44, 48). Foy used CIPC as an emulsifiable concentrate to control weeds in cotton (18). He found that there was a loss of CIPC activity due to volatilization when temperatures were over 90°F. Stark and Parochetti found that the amount of CIPC adsorbed on the soil decreased linearly with an increase in temperature (44, 51). Fang, Theisen, and Freed studied incorporated EPTC- $^{35}\text{S}$  in soil and measured vapor movement by radioactivity determination (19). At 0-3°C radioactivity remained unchanged indicating no volatility. When the temperature increased to 25°C and 35°C there was some upward movement of EPTC.

CDAA (2-chloro-N, N-diallylacetamide) (14), DNBP (4,6-dinitro-o-sec-butylphenol) (40, 26), and trifluralin (a,a,a-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine) (58) also show greater vapor loss as temperatures are elevated.

#### Moisture Effects on Volatility

Deming reported that there is a competition between water and

the herbicide CDAA for adsorption sites on the soil (14). He found<sup>7</sup> that by increasing soil moisture proportionably less CDAA was adsorbed, resulting in an increase in vapor loss. Wade observed that the sorptive capacity for ethylene dibromide on organic and clay soils decreased with increased humidity (57). CDAA (14, 50), EPTC (1, 14, 19, 25), CDEC (2-chloroallyl diethylacetamide) (25), IPC and CIPC (25, 37, 44), and DNBP (26, 41) have exhibited vapor loss as a result of increasing soil moisture.

The ability of EPTC to be more tightly adsorbed by dry soil than by wet has been reported by several workers (2, 19). Soil moisture has a marked effect on the loss of EPTC vapors (6, 22). Gray and Weieric found that during the first 15 minutes after spraying on the soil surface, 20% of the applied EPTC disappeared from dry soil, 27% from moist soil, and 44% from wet soil (22). Danielson found more loss of CIPC vapors from wet granules than from dry granules (8).

#### The Effects of Incorporation on Volatility

A common weed control practice is the incorporation of volatile herbicides in order to reduce vaporization. Mechanical incorporation of EPTC and CDEC after field application increased herbicidal activity (6, 22, 30, 47). It has been reported that dichlobenil loses one-half of its activity if there is a four hour delay before incorporation (3).

Pieczarka, Wright, and Alder found that incorporation of trifluralin increased the activity four to six times over surface spraying (46). Moist EPTC granules do not release vapors when covered with dry soil (6). However, when CIPC granules are covered with moist soil, vapor loss is observed.

Havis, Ticknar and Bobula have shown that CIPC incorporation in the soil has a tendency to reduce weed control (25). This should not be taken to mean that vapor loss is reduced. The depth of soil incorporation may reduce herbicidal activity. As the depth of soil incorporation increases there is a corresponding increase in herbicide dilution by the soil (3). Herbicide adsorption also increases with the depth of incorporation because of the increased soil surface area (46).

#### Effects of Air Movement on Volatility

As air movement increases, the persistence of volatile herbicides decreases (9, 44). Wright observed an increase in the loss of trifluralin vapor when the air flow was increased from two to six cubic feet per hour (58).

#### Mode of Action of CIPC

CIPC has been reported to inhibit cell division at the metaphase stage of mitosis (4, 16). Cytological examination has shown

chromosomal aberrations as a result of CIPC activity (16).

Swanson, Shaw, and Hughes reported that CIPC at high rates does not inhibit germination of cotton seed (52). There is a reduction in primary root length indicating that meristematic tissue is sensitive to CIPC. Since CIPC is a mitotic poison, it would be expected that all meristematic areas could be affected.

### Granular Formulation

If volatile herbicides are adsorbed on granular carriers, evaporation may be reduced. By using a proper granule carrier, surface applied herbicides can be used with a minimum of vaporization (44). Danielson studied vapor release from granular CIPC using seven different carriers (17). CIPC was very tightly held by highly adsorptive activated charcoal with no vapor or contact activity. Adsorptive clays such as calcined attapulgite resulted in the highest vapor activity (8). Granular formulation of trifluralin reduced vapor loss when compared to spray applications (58).

Post-emergence granular applications of some herbicides are considered to be safer and more effective (21, 40, 42). The herbicide granules are able to sift through heavy foliage to the moist soil where they become activated. Sprays leave a deposit on leaves resulting in crop injury and reduces the amount of spray that reaches the soil.

### Description of Dodder

Dodder is a parasitic plant belonging to the Convolvulaceae family, genus Cuscuta [Tourn.] L.(33, 45, 60). Tournefort in 1700 was the first person to describe Cuscuta (60). Yuncker in 1920 did a detailed study of the genus Cuscuta identifying and classifying 54 species (60). An excellent monographic description of dodder (Cuscuta) by Yuncker is based primarily upon floral parts:

Hypogynous, sympetalous, herbaceous parasites. Stems filiform, twining about woody or herbaceous hosts from which they obtain their nourishment by means of haustoria. Leaves reduced to small functionless scales. Flowers small, more or less cymose clustered, mostly gamosepalous; usually pentamerous (infrequently tri or tetramerous) stamens inserted in the throat, alternating with the corolla lobes; scale-like, more or less fringed or fimbriate structures present in most of the species at the base of the corolla opposite stamens; ovary two-celled, each cell containing two anatropous ovules; styles distinct or united; stigmas capitate or linear-elongated. Fruit a capsule which remains closed or opens with a distinct line or circumscission near its base; embryo acotyledonous, filiform or with enlargement at one end (60).

Species of Cuscuta can also be identified by seed size, color, and embryo position (20, 59). Large-seeded dodder (C. campestris, C. indecora) generally has seeds of 1.3 mm in diameter or more; small-seeded dodder (C. planiflora C. epithymum) has seeds of 1 mm or less in diameter (20). Gaertner found that both the anatomy of the seed coat, which consists of four layers, and the position of the spiral embryo are distinctive identification characteristics (20).

Based upon seed morphology and anatomy Gaertner has described the seeds of ten species of Cuscuta (20).

### Anatomy and Morphology

Stomata have been reported on various species of Cuscuta (55, 59). Stomata are comparatively large with at least one stoma to each several hundred epidermal cells. The stomata are sparsely scattered along the filiform stem.

The seedling shoot of Cuscuta has been reported to have no differentiated vascular tissue (28). The location of the meristematic region of the growing seedling shoot has not been well defined. The meristem has been suggested to be in the mid-section of the seedling (55).

Truscott discusses haustoria as highly modified roots of the dodder plant (56). It is well established that after a dodder seedling wraps around a suitable host, wart like appendages develop on the dodder seedling (13, 29, 53, 56). After the haustoria have started differentiating the tubercle dodder cells penetrate the host plant. The tubercle then joins with xylem of the host plant (13, 53, 56). The sieve tubes of the host and dodder plant are connected. However, this connection is probably not direct but rather by means of parenchyma cells that are sufficiently specialized to withdraw material from the host plant (56). Haustoria penetration is thought

to require enzymatic or hormonal activity by the host (29).

### Physiology

Loo reported that chlorophyll was known to exist in dodder seedlings when grown under diffuse daylight or artificial light (38). The presence of chlorophyll would suggest possible photosynthetic activity in the dodder seedling. Lane et al. found that oxygen was produced by dodder seedlings (31). Oxygen uptake was determined in the dark when samples were illuminated with a water-filtered beam of incandescent light. They found that the rate of oxygen production usually compensated for or slightly exceeded that used in respiration. Diuron (3-(3,4-dichlorophenyl)-1,1-dimethylurea), a photosynthetic inhibitor, blocked the Hill reaction of photosynthesis, but it did not stop twining of the dodder seedling (31). This would indicate that the photosynthetic activity was not important to the survival of the dodder seedling.

Dodder seedlings are sensitive to light (10, 11, 31, 61). The photocontrol of hook opening and twining of dodder seedlings have been attributed to the action of phytochrome (32, 61). Dawson found that field dodder emerged as well in total darkness as in light but seedlings did not twine about the host (10). When the dodder seedling was shaded by alfalfa, attachment was reduced more than 90%. Attached dodder shaded by alfalfa did not develop its characteristic

golden color, grew slowly and was delayed in maturity three to four weeks.

### Growth and Host Relationship

Dodder is an angiosperm; its seeds are borne within a matured ovary. One dodder plant is capable of producing hundreds of viable seeds. The seeds are gray to brown, irregularly round and have a rough surface texture (12). Usually the seedcoats are hard, which make them impermeable to water imbibition unless scarified (5, 12, 36). Dodder seeds are known to germinate after being buried in the soil for five years or longer (12).

The life cycle of the dodder seedling is specific. The various stages of development can be predicted from the time of germination until subsequent death or host attachment. Dawson (11) has reported that seedlings emerge in the greenhouse three days after planting when temperatures are 70 to 80<sup>o</sup>F and soil moisture is adequate. Within five days they will wrap around a suitable host. On the eighth day haustorial bumps appear and on the tenth day, haustoria penetration results in attachment to the host. Tronchet (55) reported that after 15 days the dodder seedling will die if it has not attached to a host plant.

Dodder emerges in a hook form similar to a bean plant. Hook opening is then dependent upon light intensity (32, 61). If light



conditions are favorable the seedling will straighten out and start rotating in a clockwise motion (36, 55). The rotation will continue up to ten or 15 days, until the seedling comes in contact with a host at which time it will wrap and attach. It is from the time of emergence until wrapping, five to ten days, that control must be accomplished (11, 54).

Dodder is adaptable to a wide range of environmental conditions. It will start germinating in early spring when the soil temperature is 60°F or higher. If moisture conditions are favorable it will germinate throughout the summer and early fall.

#### Dodder Control with CIPC

Lee, Timmons and Dawson have reported that CIPC applied to the soil killed dodder seedlings selectively in alfalfa (11, 12, 34, 35, 36, 54). Successful dodder control is not always obtained from CIPC. Dawson demonstrated that dodder control from CIPC may be incomplete if herbicide distribution, soil surface moisture, and timing of the application are not correct (11).

Dawson found that CIPC applied at six pounds per acre prior to dodder emergence gave three to six weeks of control in the greenhouse (11). CIPC often reduced but seldom prevented dodder emergence. The injured seedlings did not elongate normally or wrap on the host. Dawson also found that there was post-attachment injury to dodder plants (11). This injury was probably due to CIPC vapors being released from the CIPC granules, although proof of this assumption was lacking.

## GREENHOUSE STUDIES TO DETERMINE THE EFFECTS OF CIPC VAPORS ON DODDER SEEDLINGS

### Germination of Dodder Seeds

Mature dodder seed has a rough, hard, relatively impermeable seed coat. Under normal field conditions as little as 4% of the dodder seed will germinate at any one time (20). In order to increase the germination percentage it was necessary to treat the dodder seed with concentrated sulfuric acid.

### Materials and Methods

Two year old dodder seeds, (Cuscuta campestris and Cuscuta indecora), that had been removed from commercial lots of alfalfa seed grown in Washington, Oregon and Idaho were used in these experiments. Gaertner has demonstrated that sulfuric acid is the most expedient method of breaking the impermeable seed coat of dodder seeds (20).

Six hundred ml of concentrated sulfuric acid were added to a 2000 ml pyrex beaker. One 3/8" x 2" magnetic mixing bar was placed in the beaker. The beaker with mixing bar was placed on a Mag-mix and positioned under an exhaust hood. The Mag-mix was turned on and 100 ml of dodder seed were carefully poured into the beaker containing the sulfuric acid. The seeds were then allowed to

digest for one hour under continuous stirring from the mixing bar.

After the sulfuric acid treatment the seeds were filtered off and flushed thoroughly with tap water. This was followed by immersing the dodder seeds in a 5% sodium bicarbonate solution for 15 minutes with constant stirring. The seeds were again rinsed with tap water, then replaced in the sodium bicarbonate bath for another 15 minutes. The seeds were rinsed again with tap water, then placed on blotter paper to absorb all possible moisture. When most of the moisture was removed the seeds were placed in a flat pyrex pan covered with a plastic hair dryer. The heat from the hair dryer blower was used to complete the drying process.

After the seeds were dry 22.9 mg of Ceresan M (ethylmercury 2,3-dihydroxypropyl mercaptide + ethyl-mercury acetate) was mixed with 18.3 grams of dodder seed as a seed treatment to reduce damping-off.

Percent germination was determined by placing 100 treated dodder seeds in a 4.5" x 7/8" petri dish with appropriate germination blotters. Six petri dishes were similarly prepared. Three of the petri dishes were placed in a dark germinator at 18°C. The other three petri dishes were placed in a germinator with ten hours of light at 25°C and 14 hours of dark at 15°C. Petri dishes with untreated seeds were also placed in both dark and light germinators. The purpose was to compare treated dodder seeds with the

untreated dodder seeds. Each treatment was replicated three times. The germinated dodder seedlings were counted on the fifth day after being placed in the germinators.

### Results and Conclusions

The results of this experiment are given in Table I.

Table I. Summary of results from dodder seed treatment and germination.

	Number seeds germinated out of 100 seeds in each petri dish			Average % germination
	I	II	III	
A. Seeds treated with $H_2SO_4$ exposed only to dark environ- ment	70	56	56	60.6
Exposed to light and dark environ- ment	69	51	65	61.6
B. Seeds untreated exposed only to dark environment	5	6	4	5.0
Exposed to light and dark environ- ment	6	4	4	4.6

There was no appreciable difference in percent germination of  $H_2SO_4$  treated seeds in either the light or dark environment. There was a marked difference in treated seeds over the untreated seeds.

Not only was there a 55% increase in germination over the untreated seeds but the treated seeds all germinated within five days after being placed in the germinator. As a result of this experiment treated dodder seeds were used in all future experiments where uniform germination was required.

#### Exposure of Dodder Seedlings to CIPC Vapors in a Closed System Using Carrot Seedlings as the Host Plant

In order to study the relative effects of CIPC vapors on dodder seedlings, several experiments were conducted in the greenhouse. These involved the use of three different methods of dodder seedling exposure to CIPC vapors. The initial experiment was conducted using carrots as a host plant. Later experiments involved the use of toothpicks to act as a host substitute for the dodder seedlings.

#### Materials and Methods

In this experiment and subsequent experiments a technique similar to that developed by Meggitt (39) was used to expose dodder seedlings to CIPC vapors in a closed system. Two hundred treated dodder seeds were volumetrically seeded one cm deep in a 10 x 11.5 cm metal can filled with sterilized soil.

The soil used in all greenhouse experiments was a mixture of three parts fine sandy loam soil to one part peat moss. The results of a soil analysis before mixing with peat moss are given in Table II.

Table II. Chemical and mechanical analysis of greenhouse soil.

Soil pH	Chemical Analysis		Mechanical Analysis		
	CEC me/100g	OM %	% Sand	% Silt	% Clay
6.4	17.66	1.42	63.34	25.23	11.43

The soil used in the experiments was autoclaved using a steam sterilizing chamber. The purpose for sterilizing the soil was to reduce damping-off of the dodder seedlings and to assure consistent uniform germination of the planted dodder seeds.

Throughout the experiments greenhouse temperatures were maintained at 80°F during the day and 70°F at night.

After the dodder seeds were planted in the metal cans, a plastic bag was placed over the cans as shown in Figure 1. The purpose of the plastic bags was to reduce evaporation of soil moisture. Any irrigation that was required was accomplished by sub-irrigation.

Carrot seedlings were grown in plastic bottles 2.5 x 6 cm to be used as host plants for the emerged dodder seedlings.

On the third day after planting the dodder seedlings emerged in the classical hook stage. At that time, CIPC exposure was initiated.

To assure CIPC vapor contact to the exposed dodder seedlings a jello mold as shown in Figure 2 was used. The outside diameter



Figure 1. Dodder seedlings grown in metal can later to be exposed to CIPC vapors.

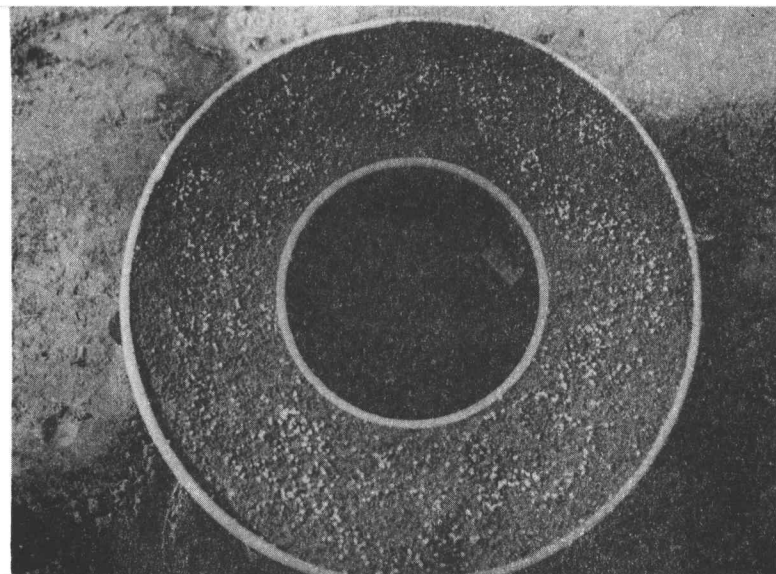


Figure 2. Jello mold with moist sterilized soil and distribution of 15 to 20 mesh attaclay CIPC granules 20% by weight.



Figure 3. Vapor trap used to expose dodder seedlings to CIPC vapors.



of the mold was 25.0 cm with the inside diameter being 10.5 cm. The mold was filled with a fine sandy loam soil and the soil moisture was brought to field capacity.

Dawson has demonstrated that granules give better dodder control than the emulsifiable concentrate or wettable powder (11). The equivalent of six pounds to the acre of 20% by weight CIPC granules, 15 to 30 mesh attaclay were applied to the moist soil surface in the jello mold. The jello molds were then placed over the metal cans containing the emerged dodder seedlings. The metal barrier between the CIPC treated soil and the soil containing the dodder seedling assured no aqueous CIPC contact to the seedlings. A plastic bag was then placed over the jello mold and can with the emerged dodder seedlings (see Figure 3). The plastic bag trapped any CIPC vapors that might be given off by the moisture activated CIPC granules. This offered a positive exposure of the vapors to the seedlings in a closed system.

The seedlings were exposed to vapors for 1, 2, 4, 8, 16, 32 and 64 hours. Each time period was replicated three times with a vapor free control for each time period.

Dodder seedlings were counted in all metal cans after the vapor chambers were removed. Also, after the chambers were removed, a plastic bottle containing a carrot seedling approximately 10 cm high was placed in each metal can to act as a host for the dodder

seedlings. Any dodder seedling that might have emerged after the vapor chamber had been removed was pulled and discarded.

Evaluation was based upon the number of dodder seedlings attached to the host plant. A seedling was considered attached if it was completely wrapped around the host plant with visible haustoria being formed. The number of attached dodder seedlings was counted and recorded 17 days after the seeding date.

### Results and Conclusions

The results of this experiment are given in Table III.

Table III. Summary of dodder seedling attachment to host when exposed to CIPC vapors in a closed system at six pounds per acre.

Hours of Exposure	No. Seedlings Emerged	No. Seedlings Attached to Host
1	109	74
2	92	32
4	100	26
8	83	13
16	78	1
32	90	0
64	111	0

Those seedlings exposed to 16 hours of CIPC vapor did not elongate normally or wrap around the host plant. Figure 4 shows

seedlings that were not exposed to CIPC vapors. The seedlings were wrapped around the host plant in a normal manner. Figure 5 shows seedlings exposed to CIPC vapors for 16 hours. The seedlings were not wrapped around the host plant. As the data in Table III indicate those seedlings exposed to CIPC vapors for 16 hours or more did not attach to the carrot host. CIPC vapors did not kill the seedling but stopped the normal process of elongation. Figure 6 shows a normal dodder seedling and a seedling that has been exposed to CIPC vapor for 16 hours.

All dodder seedlings that emerged did not necessarily attach to a host plant. This would account for the number of seedlings that did not attach in the control cans (Appendix, Table B). There was a marked decrease in seedling survival after 2, 4, and 8 hours exposure. This would indicate that there was CIPC vapor activity even when exposure was of short duration. When seedlings were exposed to vapors for 16 hours or more, 100% control was observed.

Exposure of Dodder Seedlings to CIPC Vapors  
at 32 Pounds Per Acre in a Closed System

In the preceding greenhouse experiment it was observed that at least 16 hours of seedling exposure to CIPC vapors was required for 100% control. This procedure raised the question of whether higher rates of CIPC, 32 pounds per acre, would reduce the amount of exposure time and still result in 100% control. To answer this question



Figure 4. Dodder seedlings twining about carrot host. No exposure to CIPC vapors.



Figure 5. Dodder seedling exposed to CIPC vapors for 16 hours in a closed system.

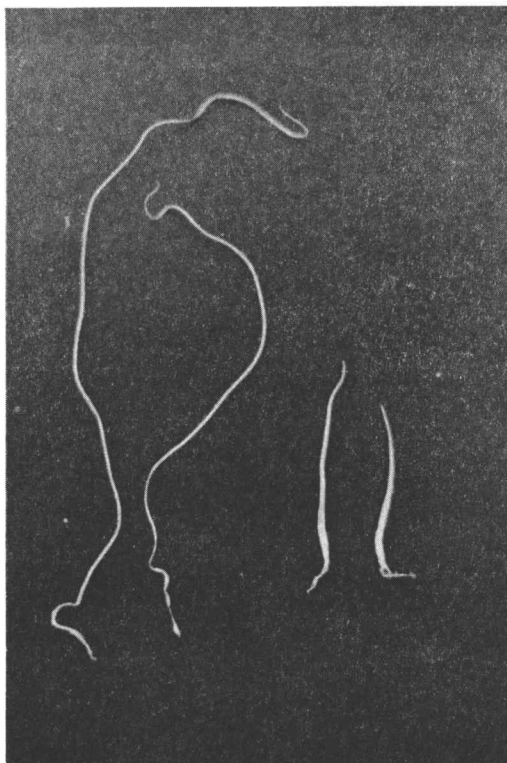


Figure 6. The two seedlings on the left were not exposed to CIPC vapors. The two seedlings on the right were exposed to CIPC vapors for 16 hours.

a similar experiment was conducted with the exception of a change in the dodder host.

### Material and Methods

The same technique used in the previous experiment was employed in this study except for the host. Carrot seedlings were satisfactory host plants but they required several weeks to grow before they could be used. In order to reduce the amount of time between experiments a substitute host was used. It was found that dodder seedlings would wrap around toothpicks and haustoria would develop. Figure 7 shows dodder seedlings wrapped around toothpicks in the same manner that they wrapped around carrot seedlings. Figure 8 actually shows the development of haustoria on the wrapped dodder seedling. Since the degree of control was measured from the time of emergence to subsequent attachment, toothpicks were found to be an adequate substitute host. Another advantage to using toothpick hosts was the number that could be used in each can. Also they could be arranged in such a manner as to obtain maximum contact surface for the emerging dodder seedlings. The toothpicks were placed 13 mm apart as indicated in Figure 9. A second experiment was conducted at 32 pounds per acre using toothpicks as a host. The same technique of exposing dodder seedlings to CIPC vapors was used.

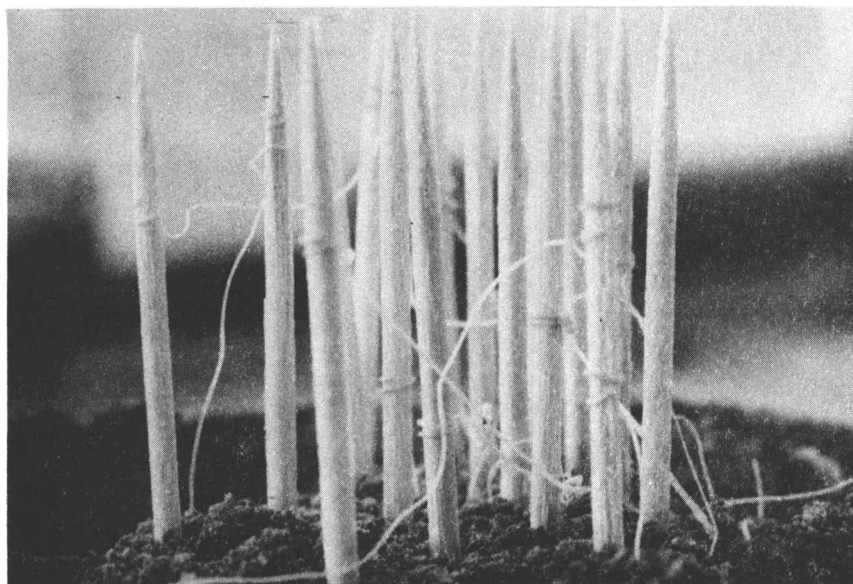


Figure 7. Dodder seedlings wrapped around the toothpick hosts.

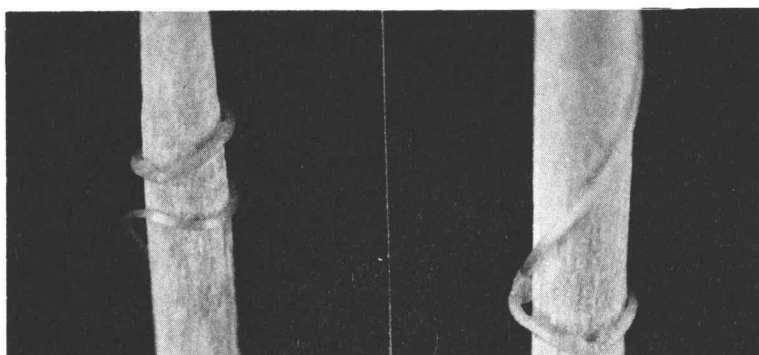


Figure 8. Dodder seedling wrapped around toothpick host showing developed haustoria.

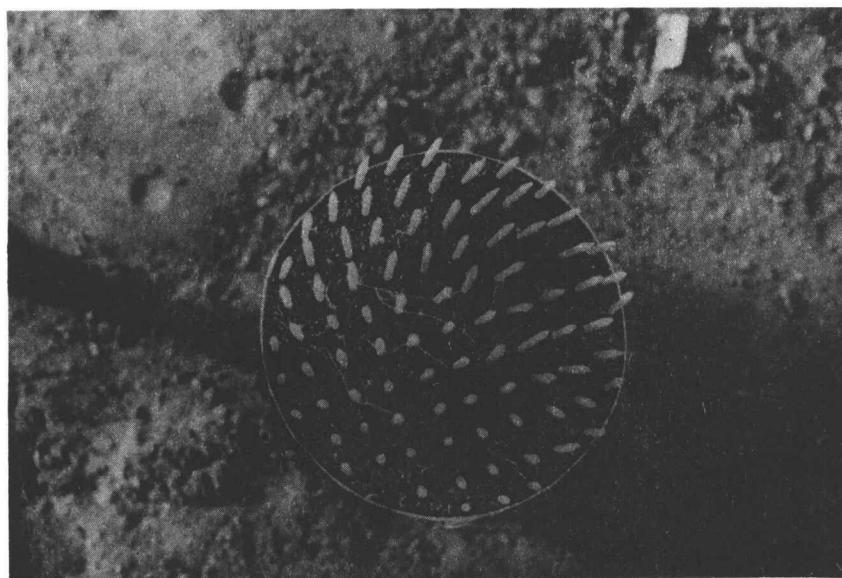


Figure 9. Toothpick hosts were spaced 13 mm apart.



## Results and Conclusions

The results of this experiment are given in Table IV.

Table IV. Summary of wrapped dodder seedling around toothpick host when exposed to CIPC vapors in a closed system at 32 pounds per acre.

Hours of Exposure	No. Seedlings Emerged	No. Seedlings Wrapped * About Toothpick Host
2	135	108
4	148	73
8	147	22
16	140	3
32	126	0
64	146	0
check	143	125

\*wrapped dodder seedlings must be wrapped around toothpick twice with well defined haustoria.

As was observed in the first experiment noticeable injury did not occur until there were at least eight hours of CIPC vapor exposure. A minimum of 16 hours of exposure was required for complete control in both experiments. This would seem to indicate that dodder seedlings must be exposed to 16 hours of CIPC vapors regardless of the rate per acre if complete control is to be obtained.

Exposure of Dodder Seedlings to CIPC Vapors  
at Six Pounds Per Acre in a Closed System  
Where the Granules Were Placed on Dry Soil

Vapor toxicity to dodder seedlings has been demonstrated in the preceding experiments. In each case 20% CIPC granules were applied to a moist soil. It has been reported that wet CIPC granules release more vapors than do dry granules (8, 11). This raised the question as to whether CIPC granules applied to a dry soil release enough vapors to be toxic to dodder seedlings.

Materials and Methods

The same technique used in the preceding experiments, vapor exposure in a closed system, was used in this experiment. The only exception was that CIPC granules were applied to air dry soil in the jello molds. The vapor exposure time was 2, 4, 8, 16 and 64 hours. Each time period was replicated four times. As in the second and third experiments, emerged dodder seedlings were counted upon removal of the vapor trap. Seventeen days from the seeding date the number of wrapped dodder seedlings with haustoria formation was counted and recorded.

Results and Conclusions

The results of this experiment are given in Table V.

Table V. Summary of wrapped dodder seedlings around toothpick host after exposure to CIPC vapors in a closed system at six pounds per acre with granules placed on dry soil.

Hours of Treatment	No. of Seedlings Emerged	No. Seedlings Wrapped About Toothpick Host
2	160	154
4	177	168
8	187	173
16	184	175
32	199	33
64	174	0
check	195	184

There was no vapor toxicity to the seedlings during the first 16 hours of exposure to CIPC vapors. Thirty two hours of exposure exhibited a marked reduction in the number of dodder seedlings wrapped about the toothpick host. Sixty four hours of exposure gave 100% control. The observations might be somewhat misleading in that the humidity in the vapor trap might build up to a high enough level to activate the CIPC granules in the later time periods. However, this experiment again displayed the fact that CIPC vapors were toxic to dodder seedlings in a closed system.

Exposure of Dodder Seedlings to  
CIPC Vapors in an Open Environment

The preceding three experiments demonstrated that CIPC vapors

were toxic to dodder seedlings in a closed system. This raised the question as to CIPC vapor effect in the open atmosphere of the greenhouse. Also, what would happen if forced air were directed over the cans containing the CIPC granules and dodder seedlings? If CIPC vapor is to be an important factor of CIPC activity in the field, then these questions must be answered. An experiment was thus conducted to expose dodder seedlings to CIPC vapors in the open atmosphere of the greenhouse.

#### Materials and Methods

It was necessary to find a method of subjecting dodder seedlings to CIPC vapors only. According to several reports, CIPC does not move readily in the soil and what movement there might be is concentrated in the top inch of soil (43, 47, 49). Dodder seedlings do not have an extensive root system. In fact the dwarf like appendages probably serve no functional purpose to the dodder seedling. Nevertheless, CIPC in a soil solution might reach the seedling and be taken up by the seedling in the aqueous form. In order to avoid this possibility, dodder seedlings were grown in 12 x 75 mm test tubes as shown in Figure 10. The test tubes were filled with sterilized soil, the same type of soil used in earlier experiments, and the moisture level was brought up to field capacity. The dodder seeds were then seeded 1 cm deep in the test tubes. The test tubes

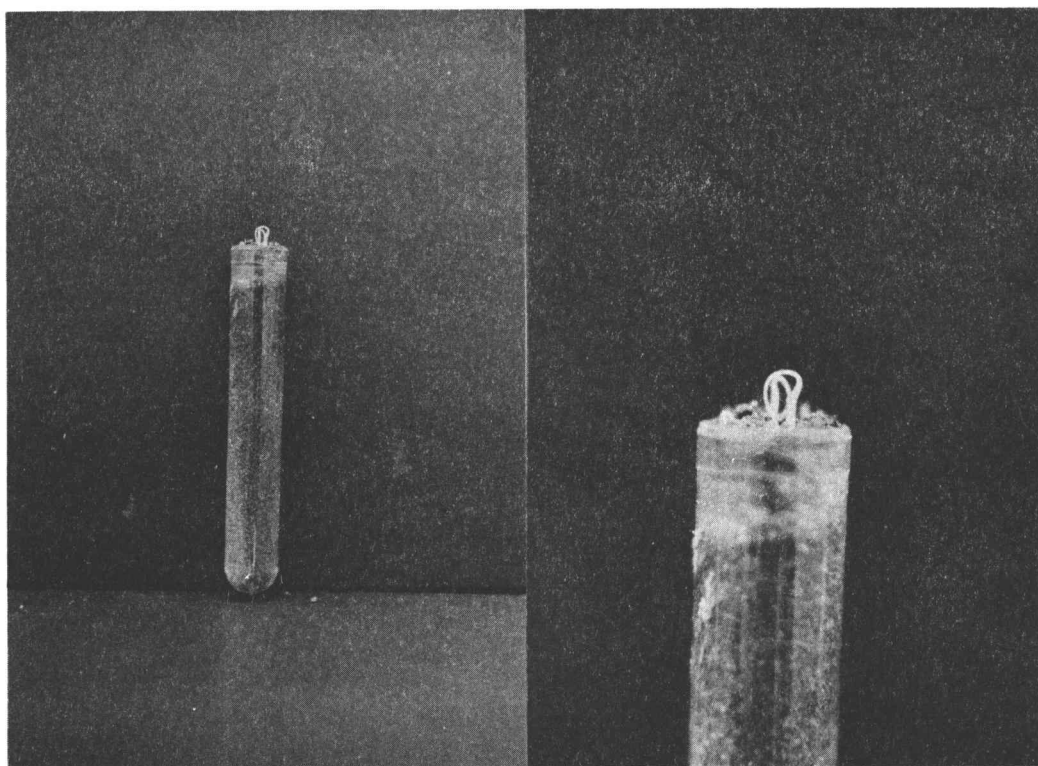


Figure 10. Hook stage dodder seedlings grown in 12 x 75 mm test tube.

were placed under a plastic covered box until the seedlings were in the hook stage of development. Sterilized soil was placed in metal cans 15 cm in diameter and 9 cm high. Ten of the test tubes containing dodder seedlings in the hook stage were placed in the metal cans. Care was taken to be sure the top of the plastic test tubes were the same level as the soil surface. Toothpicks were then placed 13 mm apart throughout each metal can to act as hosts to the dodder seedlings. CIPC 20% attaclay granules at six pounds per acre active ingredient were carefully distributed over the moist soil surface of the metal cans. Figure 11 shows the spacing of the plastic test tubes and toothpick hosts.

A series of cans similar to those in Figure 11 were set up. Dodder seeds were planted 1 cm deep in the cans in the same location as those cans containing the test tubes. CIPC 20% attaclay granules at six pounds per acre were carefully distributed over the moist soil surface of the metal cans.

The possibility that wind currents might blow the vapors away from the dodder seedlings could pose a problem under field conditions. To study this possible condition in the greenhouse a 16 inch fan was strategically located so as to allow a 10 m.p.h. velocity of wind over a third series of metal cans containing dodder seedlings and CIPC. Dodder was seeded 1 cm deep and CIPC 20% attaclay granules at six pounds per acre were evenly distributed on the moist

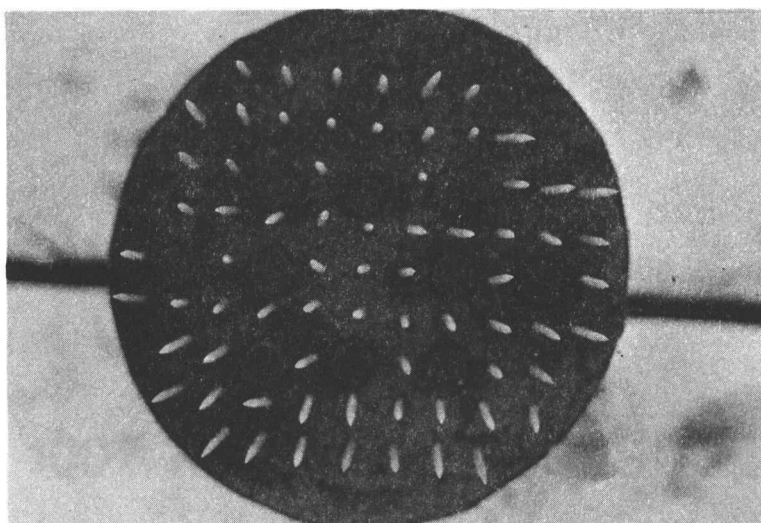


Figure 11. 12 x 75 mm test tube and toothpick host arrangement in metal can.

layer of soil. As in the other experiments toothpicks were used as the artificial host.

Each of these three methods of exposure, dodder grown in test tubes, dodder grown in cans without test tubes, air movement over dodder grown in cans without test tubes, were replicated ten times.

### Results and Conclusions

The results of this experiment are summarized in Table VI.

Table VI. Emergence and survival of dodder seedlings exposed to CIPC vapors in the open atmosphere.

Treatment	Total Number of Dodder Seedlings A Composite of Ten Reps.
CIPC at 6 lbs/acre with dodder grown in test tubes	
Emerg ed dodder seedlings	173
Wrapped dodder seedlings	0
CIPC at 6 lbs/acre with dodder grown outside test tubes	
Emerg ed dodder seedlings	208
Wrapped dodder seedlings	0
CIPC at 6 lbs/acre with dodder grown outside test tubes but with a 10 m. p. h. air motion over the top of the metal cans	
Emerg ed dodder seedlings	307
Wrapped dodder seedlings	96
Control (no CIPC treatment)	
Emerg ed dodder seedlings	272
Wrapped dodder seedlings	256

It was again demonstrated that CIPC vapors were toxic to



dodder seedlings before wrapping on a host. The results of this experiment showed this toxic effect could take place even when the vapors were released in the open atmosphere of the greenhouse. Out of 173 emerged dodder seedlings in the test tubes not one wrapped around the toothpick host after being exposed to CIPC vapors. This same condition existed when the dodder was grown without test tubes. It can be definitely established that vapors were responsible for stopping normal seedling elongation when the seedlings were grown in the test tubes. When the seedlings were grown without test tubes and aqueous CIPC could contact the seedling this assumption could not be made. In this situation it is possible that the dodder seedlings took up the CIPC in the aqueous form in addition to the vapor phase.

When dodder seedlings were exposed to both aqueous CIPC and vapors with a 10 m.p.h. simulated wind, different results were obtained. A total of 307 dodder seedlings emerged in the metal cans and after CIPC granules were applied to the soil surface 96 seedlings wrapped about the toothpick hosts. Thirty two percent of the seedlings survived which would seem to indicate that the vapors were continually being blown away thereby reducing CIPC vapor activity. This still does not prove that CIPC vapors alone are responsible for dodder control. However, it does support the hypothesis that CIPC vapors do play a role in stopping normal elongation of dodder seedlings.

Exposure of Dodder Seedlings to CIPC Vapors in an  
Open Environment Where the CIPC is in a 6 MM  
Layer of Soil Covered by 4 MM of Untreated Soil

Since CIPC does not move readily in the soil it should not go off as a vapor if covered by soil. The question then arises whether CIPC vapors might penetrate 4 mm of untreated soil and still cause injury to dodder seedlings grown in a test tube. What would happen to dodder seedlings that germinate below the 6 mm CIPC band? Along this same line of thought would a 4 mm top layer of activated carbon adsorb all possible vapors released in a 6 mm band of treated soil and thereby allow dodder seedlings in test tubes to wrap normally about a toothpick host?

To answer these questions two experiments were set up under greenhouse conditions.

Materials and Methods

The technique developed to study the foregoing questions was based upon using CIPC as an emulsifiable concentrate thoroughly mixed in a sandy loam soil. A 6 mm band of CIPC treated soil was placed 4 mm below the soil surface. Figure 12 shows the method used in placing the test tubes, CIPC treated band of soil, and toothpicks.

The following experiments were replicated five times.

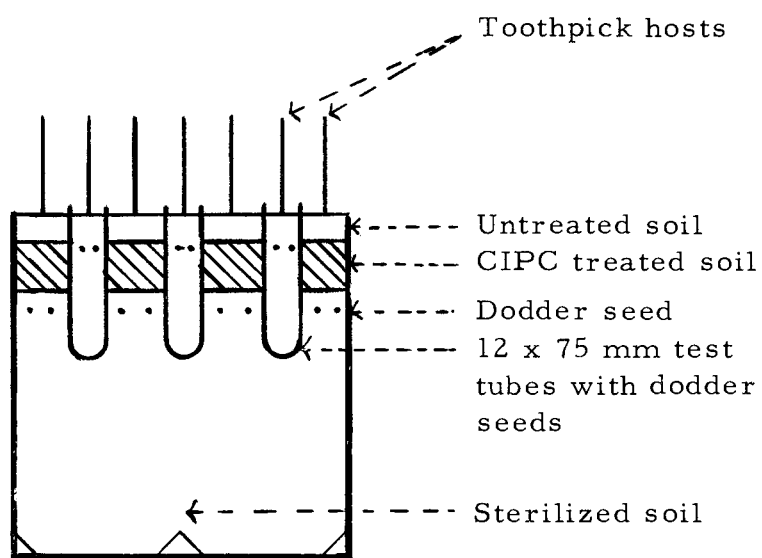


Figure 12. Technique used to expose dodder seedling to a subterranean layer of CIPC mixed with the soil.

Approximately 200 treated dodder seeds were seeded at a depth of 13 mm in a metal can containing sterilized soil. Each metal can was 15 cm in diameter and 9 cm high. The seeds were then covered by 4 mm of untreated sterilized soil. The next layer of soil was treated with CIPC at 72 ppm (approximately six pounds per acre). The uniform mixing of soil and herbicide was accomplished by mixing the air dried sterilized soil with water and CIPC solution in a mechanical soil blender. This brought the CIPC treated soil moisture level to 18%. The treated soil was immediately placed on top of the untreated soil covering the dodder seed. The 6 mm layer of CIPC treated soil was then covered with 4 mm of untreated sterilized soil. Figure 12 shows the layering technique used in this study.

Ten 12 x 75 mm plugs of soil were removed from each metal can so that the 12 x 75 mm test tubes could be placed in the metal cans containing the layered soil. Toothpick hosts were then placed 13 mm apart throughout each metal can.

It was thought that if the dodder seedlings in the test tubes were injured then it would have to be from CIPC vapors that had penetrated the surface layer of untreated soil. If the test tube seedlings were not injured but the dodder seedlings grown below the CIPC layer were injured this might indicate injury due to aqueous uptake of CIPC by the seedlings. This did not discount the possibility that CIPC vapors in the soil were taken up by the emerging seedlings.

Activated carbon will strongly adsorb CIPC (8). By using a 4 mm layer of activated carbon over the CIPC band it should adsorb any vapor that might escape to the soil surface and atmosphere.

Another question that seemed pertinent to CIPC exposure in the soil was the difference in CIPC concentration in the soil. A similar experiment was carried out to determine if concentration made any difference as to CIPC volatility in the soil.

All experiments were sub-irrigated.

### Results and Conclusions

The results of dodder seedling emergence and survival when exposed to CIPC as an emulsifiable concentrate banded below the soil surface are summarized in Table VII.

CIPC vapors were being released from the treated band of soil as demonstrated by the fact that only four seedlings wrapped on the toothpicks from the 188 seedlings exposed to vapors in the test tubes. A total of 70 seedlings were able to grow through the 6 mm band of CIPC treated soil. However, not one of these seedlings wrapped around a toothpick host. Figure 13 shows the difference between seedlings exposed to the CIPC band and those that were not exposed. Seedlings B show the normal stunting with a marked thickening of the seedling stem. Seedlings A emerged normally; 14 days from seeding they had wrapped and formed haustoria.

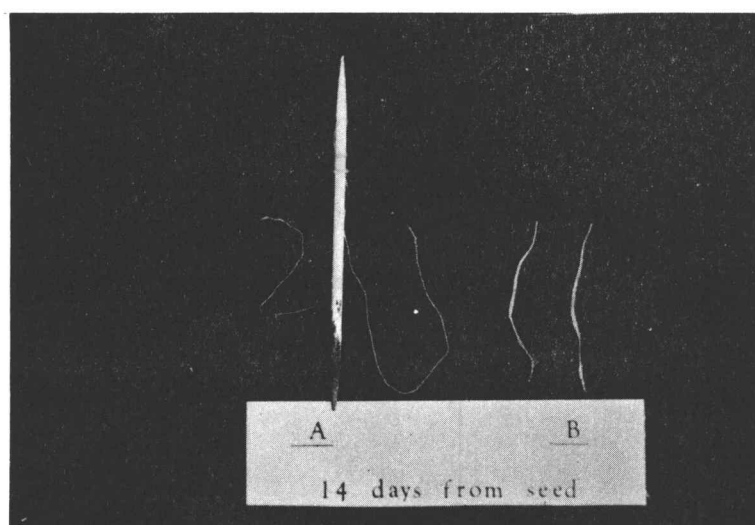


Figure 13. A. Seedlings emerged normally and wrapped around the toothpick host. B. Seedlings exposed to CIPC showed stunting with a thickening of the stem.

Table VII. Summary of dodder seedling emergence and survival where seedlings were exposed to CIPC as an emulsifiable concentrate in a 6 mm band of soil covered by 4 mm of untreated soil.

Treatment	Total Number of Dodder Seedlings
Dodder seeded 4 mm below the 6 mm band of CIPC treated soil	
Emerged dodder seedlings in metal cans	70
Wrapped * dodder seedlings from metal cans	0
Emerged dodder seedlings in 12 x 75 mm test tubes	188
Wrapped dodder seedlings from 12 x 75 mm test tubes	4
Control (No CIPC treatment)	
Emerged dodder seedlings in metal cans	229
Wrapped dodder seedlings from metal cans	217
Emerged dodder seedlings from 12 x 75 mm test tubes	193
Wrapped dodder seedlings from 12 x 75 mm test tubes	183

\*wrapped dodder seedlings must be wrapped around toothpicks twice with well defined haustoria.

Further evidence of CIPC activity when applied as an emulsifiable concentrate in subterranean layered soil was reported in Table VIII.

The data from Table VIII again demonstrated that CIPC vapors are released from the subterranean soil at a concentration high enough to injure the seedlings in the test tubes. As the CIPC concentration decreased from 36 ppm to 6 ppm there was a noticeable decrease in the toxic vapor effect on the test tube seedlings. At 36 ppm only ten seedlings wrapped from 111 that emerged in the test

Table VIII. Summary of dodder seedling emergence and survival where seedlings were exposed to different concentrations of CIPC as an emulsifiable concentrate in a subterranean band of soil.

Treatment	Total Number of Dodder Seedlings
<u>Dodder seeded 4 mm below the 6 mm band of CIPC treated soil</u>	
CIPC 36 ppm	
Emerg ed dodder seedlings in cans	49
Dodder seedlings wrapped about toothpicks	0
Emerg ed dodder seedlings in test tubes	111
Dodder seedlings wrapped about toothpicks	10
CIPC 12 ppm	
Emerg ed dodder seedlings in cans	110
Dodder seedlings wrapped about toothpicks	0
Emerg ed dodder seedlings in test tubes	110
Dodder seedlings wrapped about toothpicks	39
CIPC 6 ppm	
Emerg ed dodder seedlings in cans	108
Dodder seedlings wrapped about toothpicks	0
Emerg ed dodder seedlings in test tubes	102
Dodder seedlings wrapped about toothpicks	57
<u>Control</u>	
Emerg ed dodder seedlings in cans	84
Dodder seedlings wrapped about toothpicks	80
Emerg ed dodder seedlings in test tubes	110
Dodder seedlings wrapped about toothpicks	102



tubes. When the CIPC concentration was reduced to 6 ppm, 57 seedlings wrapped from 102 that emerged in the test tubes. These data seem to indicate that at even extremely low concentration, vapors are released from subterranean treated soil.

Regardless of the concentration, 6 ppm or 36 ppm, those seedlings that emerged through the 6 mm band of treated soil did not wrap around the toothpick host. As the concentration level decreased from 36 ppm to 6 ppm more seedlings emerged through the band. At 36 ppm a total of only 49 seedlings emerged through the treated layer, whereas, at 12 ppm, 110 seedlings emerged through the treated band. This difference in CIPC activity in the soil might be explained by the fact that there was less CIPC available for uptake by the seedling through either the aqueous or vapor phase. At low rates of 6 or 12 ppm, most of the CIPC molecules remained tightly adsorbed to the soil organic matter. However, those molecules that were released entered the vapor phase and diffused to the soil surface as demonstrated by the number of seedlings injured in the test tubes.

The CIPC vapor toxicity to seedlings growing in the test tubes could be stopped by adding a 4 mm layer of activated carbon above the 6 mm layer of CIPC treated soil. Table IX summarizes the results of such an experiment where activated carbon was layered on the treated subterranean soil.

Table IX. Summary of dodder seedling emergence and survival where seedlings were exposed to CIPC as an emulsifiable concentrate in a 6 mm band of soil covered by 4 mm of activated carbon.

Treatment	Total Number of Dodder Seedlings
Dodder seeded 4 mm below the 6 mm band of CIPC treated soil which was covered by a 4 mm layer of activated carbon	
Emerged dodder seedlings in cans	14
Dodder seedlings wrapped about toothpicks	0
Emerged dodder seedlings in test tubes	131
Dodder seedlings wrapped about toothpicks	122

By using activated carbon as a layer over the subterranean layer of CIPC treated soil almost 100% of the test tube seedlings wrapped around the toothpick hosts. The vapors being released by the subterranean treated soil were unable to penetrate the layer of activated carbon, probably because of adsorption of the vapors by the carbon. Those seedlings that emerged through the treated layer of soil plus the layer of activated carbon did not wrap around the toothpick hosts.

This still does not give proof as to whether vapors or aqueous solution was causing the injury to the seedling that must emerge through the treated layer. It was reasonable to assume that both vapors and aqueous solution contact the seedling in the subterranean layer. Still some of the seedlings were able to penetrate this layer

and grow to a height of 15 mm before normal elongation stopped.

More research is needed to determine the exact site of uptake by the dodder seedling. Also, a technique is needed to separate the aqueous phase from the vapor phase.

## A FIELD STUDY ON THE EFFECTS OF CIPC VAPORS ON DODDER SEEDLINGS

The previous greenhouse experiments demonstrated the phytotoxic effects of CIPC vapors on dodder seedlings. In the greenhouse studies, environmental variables were controlled, i. e., temperature was maintained at 80°F during the day and 70°F at night, air movement was introduced only when desired, and moisture was only by sub-irrigation. The question that must be answered, if the information discovered in the greenhouse is to be of practical value, is whether the same results could be duplicated under field conditions. In order to answer the question, an experiment was carried out at the Hyslop Farm five miles northwest of Corvallis, Oregon.

### Materials and Methods

Dodder is a common pest in alfalfa, especially seed alfalfa. Since alfalfa makes an excellent host plant this plant species was used in the field experiment.

The alfalfa was clipped at two different heights, 5.1 cm and 15 cm. The purpose for the two different heights was to see whether the long stubble would have a tendency to hold the vapors for a longer period of time than short stubble. If more dodder seedlings attached in the short stubble than the long stubble, then it could be assumed that a canopy of alfalfa would help to enhance CIPC vapor activity.

CIPC 20% attaclay granules using six pounds active ingredient to the acre were uniformly distributed over an area 20 feet by 30 feet in each of the clipped areas using a Barber spreader. One day prior to CIPC application the entire alfalfa field was sprinkler irrigated with three inches of water. This assured satisfactory soil moisture for optimum dodder control with CIPC.

Dodder seedlings were grown in the greenhouse in test tubes similar to Figure 10. Fourteen test tubes containing at least ten dodder seedlings in the hook stage were placed at random in both of the different height plots. Each time that dodder seedlings in the test tubes were placed in the treated plots seven test tubes containing at least ten dodder seedlings in the hook stage were placed in each of two untreated control plots. Care was taken to place the test tubes so that they were close to an alfalfa host.

Test tubes with dodder seedlings at hook stage were set out the day after the CIPC application. The following sequence was followed in putting out dodder seedlings grown in test tubes. A complete series of test tubes, 14 in each of the two treated plots plus seven in each of the two untreated control plots were set out every day for the first six days; then at intervals of three days for the remainder of the experimental period. This length of time consisted of 31 days. The alfalfa height at the end of 19 days was 35 mm in the short stubble plot and 37.5 cm in the long stubble plot. At the end of the 31 day

period, the alfalfa was 61 cm high in both plots.

Fifteen days after the test tubes containing the dodder seedlings were set out, seedlings were counted and recorded. Attachment was determined by complete wrapping of the seedling with well developed haustoria. Color of the seedling was also used as a method for determining seedling survival. If the attached seedling was orange then the seedling was assumed firmly attached to the alfalfa host.

During the length of the experiment, maximum and minimum daily temperatures, daily precipitation, and mean daily wind velocity and direction were obtained from the Hyslop weather station.

The field containing the alfalfa plots was irrigated two times during the experiment by sprinkler irrigation. A total of eight inches of water was applied to the alfalfa.

### Results and Conclusions

During the experimental period, the maximum daily temperature was 82.7°F, the minimum 50.6°F. The average wind velocity during this period was 5.9 m. p. h. primarily from the north. Detailed daily temperatures and wind velocity can be found in Table I in the Appendix.

The results of seedling emergence and survival are given in Table X and Table XI.

Dawson reported that dodder control in seed alfalfa must be

complete or nearly so (11). Even though there might be a statistically significant difference between treated and untreated plots, this difference was usually not great enough to give satisfactory dodder control. At least a 95 to 100% reduction in wrapped or attached dodder seedlings was considered the criteria of control rather than a statistically significant difference (11). This same standard was used in evaluating the studies conducted in this thesis.

Table X. Summary of dodder seedling emergence and survival where seedlings were exposed to CIPC vapors under field conditions in an alfalfa stubble height of 5.1 cm.

Date seedlings were placed in field		Total emerged dodder seedlings in test tubes		Total attached dodder seedlings on alfalfa	
		Treated Plot	Check Plot	Treated Plot	Check Plot
July	21	22	9	0	6
	22	27	13	1	7
	23	26	12	0	8
	24	23	10	0	7
	25	24	13	1	9
	26	20	10	0	6
	30	21	12	5	10
August	2	21	10	1	4
	5	20	11	0	4
	8	21	10	0	3
	11	23	9	1	2
	14	22	10	0	1
	17	21	11	0	0
	20	23	12	0	1
	Total	314	152	9	68

Table XI. Summary of dodder seedling emergence and survival where seedlings were exposed to CIPC vapors under field conditions in an alfalfa stubble height of 15 cm.

Date seedlings were placed in field		Total emerged dodder seedlings in test tubes		Total attached dodder seedlings on alfalfa	
		Treated Plot	Check Plot	Treated Plot	Check Plot
July	21	25	10	0	7
	22	22	13	0	6
	23	23	11	0	7
	24	23	13	0	4
	25	29	11	1	5
	26	22	12	2	3
	30	21	10	0	5
August	2	23	9	0	1
	5	22	10	0	0
	8	20	11	0	2
	11	22	10	0	3
	14	22	9	0	0
	17	20	11	0	2
	20	23	10	0	0
Total		317	150	3	45

CIPC vapors were phytotoxic to dodder seedlings under field conditions. Essentially complete control of dodder seedlings was obtained in either the 5.1 cm or 15 cm alfalfa stubble plots.

Ninety six percent of the emerged dodder seedlings failed to attach in the 5.1 cm stubble plot. In the 15 cm plot 99% of the emerged dodder seedlings did not attach to the alfalfa host.



The mortality of dodder seedlings in the untreated checks seemed to be rather high. In the 5.1 cm stubble plot 68 seedlings attached to the host from 152 seedlings that emerged. A similar mortality rate was observed in the 15 cm stubble plot. A total of 150 seedlings emerged from the test tubes but only 45 attached to the alfalfa host.

This perhaps can be explained on the basis of shading from the alfalfa plants. Dodder seedlings are reported to be light dependent (38, 55). Dawson also found that shade from tall growing alfalfa suppressed dodder (15). This same effect of dodder suppression was observed in this study when the alfalfa grew high enough to form a canopy over the emerging dodder seedlings; attachment was greatly reduced or stopped entirely. This reduction in attachment can be seen from the data presented in Table X and XI. Shading had a greater effect in the 15 cm stubble plot than in the shorter 5.1 cm stubble plot.

## DISCUSSION

In order to obtain a high degree of uniform germination of dodder seeds it was necessary to scarify the seeds before planting. By treating the seeds with concentrated sulfuric acid the percent germination was increased by approximately 55%. However, the seedlings had a tendency to damp-off when germinated in unsterilized soil. The seeds were treated with Ceresan-M and germinated in sterilized soil. By using these two methods the incidence of damping-off of dodder seedlings was controlled.

The use of sterilized soil in a persistence trial to measure CIPC vapor loss over a long period of time could lead to erroneous conclusions. The destruction of micro-organisms by soil sterilization would increase the persistence of CIPC. Microbial degradation of CIPC has been found to be an important method of decomposition in the soil. However, in the short time periods of CIPC exposure used for the greenhouse experiments, microbial degradation was not considered to be an important factor.

Under field conditions only a small fraction of the dodder seeds germinate at any one time. It is possible, if environmental conditions are favorable, for dodder seeds to germinate throughout the summer months. Therefore, a chemical control program must, by necessity, give 100% control during the period of time dodder

seedlings might attach and survive on a suitable host plant.

CIPC has been found to control dodder for four to five weeks. This time period is sufficient to obtain adequate dodder control in seed alfalfa. The degree of control is based upon such factors as soil moisture, CIPC granule distribution, wind velocity and temperature. Sometimes control is sporadic for reasons that are not well defined.

Results obtained from these studies indicate that CIPC vapors are important in stopping the normal elongation of dodder seedlings. In order to obtain 100% dodder control, 16 hours of exposure to CIPC vapor were required whether the rate was six pounds per acre or 32 pounds per acre. It would be misleading to say that different rates of CIPC influenced the time of exposure required to give 100% dodder control. In a closed system the vapors would not be able to escape into the atmosphere. As a result a concentration gradient would build up regardless of the CIPC rate.

When CIPC granules were applied to a dry soil in a closed system 64 hours of vapor exposure was required to give 100% dodder control. There are two possible reasons for this increase in time of exposure. First, vapors were being released from the dry CIPC granules, but at such a slow rate that 64 hours were required for the vapor concentration to reach a level that would be lethal to the dodder seedlings. Second, as time elapsed in the closed system, moisture

evaporation from the soil took place in the cans containing the dodder seedlings. As the humidity increased in the closed system the CIPC granules on the dry soil were activated by water absorption from the atmospheric moisture confined in the vapor chamber. CIPC vapors were then released and absorbed by the dodder seedlings.

It is not clear how vapors enter into the dodder seedling. There are two possible routes, through the stomata on the stem or direct penetration of the stem cuticle. By using radioactive CIPC, the method of vapor uptake by the dodder seedling could probably be determined.

CIPC vapors were also found to be toxic to dodder seedlings in the open atmosphere. Plastic test tubes filled with sterilized soil were seeded with dodder seeds. When the seedlings were in the hook stage of growth they were exposed to CIPC vapors in the open atmosphere. In every case 100% dodder control was obtained, again demonstrating the phytotoxic properties of CIPC vapors to dodder seedlings.

CIPC in soil solution always exhibited toxicity to dodder seedlings growing in the same soil. It has been demonstrated that vapors are toxic to dodder seedlings, but it is not known how CIPC enters the seedling in the soil.

In the soil, CIPC is present as a vapor and also as an aqueous solution. No method was found to separate the CIPC vapors from

aqueous CIPC when applied to the soil. The emerging dodder seedling and the basal part of the dodder seedling would be exposed to vapors in the soil as well as CIPC in soil solution. To say that only vapors affect dodder seedlings would not be true. If a method could be developed to restrict any possible CIPC vapor uptake in the soil or atmosphere, then definite conclusions could be made as to the primary mode of CIPC uptake by the dodder seedlings. If the seedlings all wrapped around a host plant when exposed only to aqueous CIPC then it must be the vapors that are entering the seedling through the stomata or cuticle.

It is possible to expose only the upper part of the seedling to CIPC vapors. The results of both greenhouse and field experiments have demonstrated that CIPC vapors will stop normal elongation of dodder seedlings when the seedlings are isolated from the soil containing the CIPC.

When CIPC was mixed with soil and covered with a 4 mm layer of untreated soil, the vapors were able to penetrate the untreated soil. This was demonstrated by the complete control of dodder seedlings grown in test tubes and exposed to CIPC vapors from the CIPC-treated subterranean soil. The vapors being released from the treated soil were completely stopped by covering this treated soil with 4 mm of activated carbon. The carbon adsorbed all the CIPC molecules before they could reach the open atmosphere. As a

result, most of the test tube dodder seedlings wrapped around toothpick hosts. Evidence of the diffusion of CIPC vapors to the soil surface where they can become toxic to dodder seedlings suggests that vapors can also diffuse in the soil and be absorbed by emerging seedlings.

Alfalfa seed production areas in the United States are known to have consistently strong winds, 15 to 25 m.p.h., in the spring and early summer. These areas do not always get good dodder control with CIPC. The reason for poor control could very well be the result of the CIPC vapors being blown away before they can exert their phytotoxic properties on dodder seedlings. In areas where wind is a problem, planting trees for wind breaks or planting row alfalfa opposite the prevailing wind current might reduce CIPC vapor loss and result in better dodder control.

The results of this thesis research suggests that CIPC vapor activity may be the primary factor in dodder control.

## SUMMARY

Studies were conducted to determine the effect of CIPC vapors on dodder seedlings prior to attachment or wrapping. Exposure of dodder seedlings to CIPC vapors for 2, 4, 8, 16, 32, and 64 hours in a closed system was studied. The effects of soil-incorporated CIPC as it related to volatility was investigated. The following results were obtained:

1. CIPC as 15 to 30 mesh attaclay granules containing 20% CIPC was applied to moist and dry soil at different rates in a closed vapor trap system. It was found that a minimum of 16 hours of exposure to CIPC vapors was necessary to obtain 95 to 100% dodder control when the granules were applied to a moist soil. Sixty four hours of exposure were required when granules were placed on a dry soil.
2. A technique of using toothpicks as a substitute host plant was found to be very effective.
3. CIPC vapor exposure of dodder seedlings in the hook stage grown in test tubes was found to be an effective method of separating CIPC vapors from CIPC in soil solution.
4. Dodder seedlings were found to be injured by CIPC vapors when grown in the open greenhouse.
5. CIPC vapors also produced phytotoxic properties under field conditions. This was demonstrated by an experiment in which seedlings grown in test tubes were exposed to CIPC vapors in an alfalfa field trial.

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## APPENDIX

## APPENDIX

Tables in this section have been presented  
as condensed summaries in the main text.

Table A. Emergence and survival of dodder seedlings exposed to CIPC vapors in a closed system at six pounds per acre of CIPC on moist soil.

Hours of Exposure	No. of Seedlings Emerged				No. of Seedlings Attached to Host (carrots)			
	I	II	III	Total	I	II	III	Total
1	40	32	37	109	22	27	25	74
2	23	37	32	92	10	13	9	32
4	26	39	35	100	9	10	7	26
8	31	22	30	83	6	4	3	13
16	24	25	29	78	1	0	0	1
32	37	25	28	90	0	0	0	0
64	41	29	31	111	0	0	0	0
Hours of Exposure	Check				Check			
1	29				24			
2	31				23			
4	39				31			
8	29				23			
16	27				20			
32	33				24			
64	30				27			



Table B. Emergence and survival of dodder seedlings exposed to CIPC vapors in a closed system at 32 pounds per acre.

Hours of Treatment	No. Seedlings Emerged					No. Seedlings Attached to Host (Toothpicks)				
	I	II	III	IV	Total	I	II	III	IV	Total
2	37	39	29	30	135	34	30	21	23	108
4	36	37	37	38	148	23	15	22	13	73
8	30	41	34	42	147	8	2	2	10	22
16	46	27	35	32	140	0	0	1	2	3
32	40	25	31	30	126	0	0	0	0	0
64	31	32	40	43	146	0	0	0	0	0
Check	33	40	40	30	143	29	38	33	25	125

Table C. Emergence and survival of dodder seedlings exposed to CIPC vapors in a closed system at six pounds per acre on dry soil.

Hours of Treatment	No. Seedlings Emerged					No. Seedlings Attached to Host (Toothpicks)				
	I	II	III	IV	Total	I	II	III	IV	Total
2	46	36	37	41	160	44	34	35	41	154
4	46	33	55	43	177	41	33	54	40	168
8	36	37	59	55	187	36	30	54	53	173
16	35	51	40	58	184	32	47	39	57	175
32	55	53	48	43	199	11	11	7	4	33
64	45	51	38	40	174	0	0	0	0	0
Check	40	46	58	51	195	37	44	55	48	184

Table D. Emergence and survival of dodder seedlings exposed to CIPC vapor and to CIPC in soil solution at six pounds per acre in the greenhouse atmosphere.

Treatment	Number of Seedlings/Rep										Total
	I	II	III	IV	V	VI	VII	VIII	IX	X	
CIPC at 6 lb/acre with dodder grown in test tubes											
Emerged dodder seedlings	21	24	21	18	14	21	15	10	12	17	173
Attached dodder seedlings	0	0	0	0	0	0	0	0	0	0	0
CIPC at 6 lbs/acre without test tubes											
Emerged dodder seedlings	19	26	23	20	24	17	14	21	23	21	208
Wrapped dodder seedlings	0	0	0	0	0	0	0	0	0	0	0
CIPC at 6 lbs/acre without test tubes but with fan (air movement 10 mph)											
Emerged dodder seedlings	32	27	43	28	16	25	30	32	42	32	307
Wrapped dodder seedlings	9	4	15	9	4	5	10	14	12	14	96
Control No Treatment											
Emerged dodder seedlings	31	32	29	24	22	23	29	28	24	30	272
Wrapped dodder seedlings	31	31	20	24	22	22	28	28	23	27	256

Table E. Emergence and survival of dodder seedlings where seedlings were exposed to CIPC as an emulsifiable concentrate mixed in a 6 mm layer of soil covered by 4 mm of untreated soil.

Treatment	No. of Dodder Seedlings					Total
	I	II	III	IV	V	
Dodder seeded 4 mm below the 6 mm band of CIPC treated soil						
Emerged dodder seedlings in metal cans	14	13	15	13	15	70
Dodder seedlings wrapped about toothpicks in metal cans	0	0	0	0	0	0
Emerged dodder seedlings in 12 x 75 mm test tubes	37	37	46	39	29	188
Dodder seedlings wrapped about toothpicks in 12 x 75 mm test tubes	0	1	1	0	2	4
Nontreated Cans						
Emerged dodder seedlings in metal cans	48	41	52	49	39	229
Dodder seedlings wrapped about toothpicks	45	41	49	47	35	217
Emerged dodder seedlings in 12 x 75 mm test tubes	47	29	41	39	37	193
Dodder seedlings wrapped about toothpicks in 12 x 75 mm test tubes	44	25	40	38	36	183

Table F. Emergence and survival of dodder seedlings where seedlings were exposed to CIPC as an emulsifiable concentrate mixed in a 6 mm layer of soil covered by 4 mm layer of untreated soil.

Treatment	No. of Dodder Seedlings			Total
	I	II	III	
Dodder seeded 4 mm below the 6 mm band of CIPC treated soil				
CIPC 36 ppm				
Dodder seedlings in can exposed to vapors	10	19	20	49
Dodder seedlings wrapped about toothpicks	0	0	0	0
Dodder seedlings in test tubes exposed to vapors	37	39	35	111
Dodder seedlings wrapped about toothpicks	2	5	3	10
CIPC 12 ppm				
Dodder seedlings in can exposed to vapors	42	37	31	110
Dodder seedlings wrapped about toothpicks	0	0	0	0
Dodder seedlings in test tubes exposed to vapors	29	31	50	110
Dodder seedlings wrapped about toothpicks	10	11	18	39
CIPC 6 ppm				
Dodder seedlings in can exposed to vapors	25	42	41	108
Dodder seedlings wrapped about toothpicks	0	0	0	0
Dodder seedlings in test tubes exposed to vapors	27	32	43	102
Dodder seedlings wrapped about toothpicks	14	24	19	57
Control No CIPC Treated soil				
Dodder seedlings in can not exposed to vapors	25	29	30	84
Dodder seedlings wrapped about toothpicks	25	27	28	80
Dodder seedlings in test tubes not exposed to vapors	32	36	42	110
Dodder seedlings wrapped about toothpicks	32	33	37	102

Table G. Emergence and survival of dodder seedlings where seedlings were exposed to CIPC as an emulsifiable concentrate mixed in a 6 mm layer of soil covered by 4 mm of activated carbon.

Treatment	No. of Dodder Seedlings			
	I	II	III	Total
Dodder seeded 4 mm below the 6 mm band of CIPC treated soil which was covered by 4 mm of activated carbon				
Emerged dodder seedlings in can	6	5	3	14
Dodder seedlings wrapped about toothpicks	0	0	0	0
Emerged dodder seedlings in test tube	43	43	45	131
Dodder seedlings wrapped about toothpicks	39	40	43	122

Table H. Emergence and survival of dodder seedlings grown in a test tube and exposed to CIPC vapors under field conditions. Seedling placed adjacent to 5.1 cm high alfalfa stubble.

Date of Initial Exposure		Emergenced Dodder Seedlings in Test Tube				Attached Dodder Seedlings to Alfalfa			
		I	II	Total	Check	I	II	Total	Check
July	21	10	12	22	9	0	0	0	6
	22	13	14	27	13	0	1	1	7
	23	14	12	26	12	0	0	0	8
	24	10	13	23	10	0	0	0	7
	25	10	14	24	13	1	0	1	9
	26	10	10	20	10	0	0	0	6
August	30	11	10	21	12	3	2	5	10
	2	10	11	21	10	0	1	1	4
	5	10	10	20	11	0	0	0	4
	8	11	10	21	10	0	0	0	3
	11	12	11	23	9	0	1	1	2
	14	11	11	22	10	0	0	0	1
	17	10	11	21	11	1	0	0	0
	20	11	12	23	12	0	0	0	1
		Total	314	152	Total		9	68	

Table I. Emergence and survival of dodder seedlings grown in a test tube and exposed to CIPC vapors under field conditions. Seedlings placed adjacent to 15 cm high alfalfa stubble.

Date of Initial Exposure		Emergenced Dodder Seedlings in Test Tube				Attached Dodder Seedlings to Alfalfa			
		I	II	Total	Check	I	II	Total	Check
July	21	11	14	25	10	0	0	0	7
	22	10	12	22	13	0	0	0	6
	23	10	13	23	11	0	0	0	7
	24	10	13	23	13	0	0	0	4
	25	14	15	29	11	0	1	1	5
	26	12	10	22	12	1	1	2	3
August	30	11	10	21	10	0	0	0	5
	2	13	10	23	9	0	0	0	1
	5	11	11	22	10	0	0	0	0
	8	10	10	20	11	0	0	0	2
	11	11	11	22	10	0	0	0	3
	14	12	10	22	9	0	0	0	0
	17	10	10	20	11	0	0	0	2
	20	10	13	23	10	0	0	0	0
		Total	317	150	Total		3	45	

Table J. Climatological conditions at Hyslop Farm from July 21, 1966 to August 30, 1966.

Date	Air Temperature F <sup>o</sup>		Precipitation	Wind Velocity		
	Max	Min	Rain-Inches	Mean Daily Ave. Speed mph	Ave. Direction	
July	21	79	51	0	6	N
	22	89	48	0	5	N
	23	89	48	0	5	W
	24	85	56	0	5	W
	25	72	42	0	5	N
	26	78	52	0	8	N
	27	84	54	0	6	N
	28	90	48	0	5	N
	29	90	52	0	8	N
	30	94	48	0	5	N
	31	85	52	0	6	N
August	1	84	54	0	7	N
	2	88	55	0	6	N
	3	92	50	0	3	N
	4	89	56	0	5	N
	5	82	54	0	8	N
	6	83	53	0	7	N
	7	86	52	0	4	N
	8	90	48	0	3	N
	9	80	54	0	5	N
	10	81	53	0	8	N
	11	81	50	0	9	N
	12	81	48	0	5	N
	13	86	53	0	5	N
	14	80	54	0	8	N
	15	84	55	0	7	N
	16	87	52	0	10	N
	17	82	49	0	8	N
	18	85	46	0	5	N
	19	86	45	0	8	N
	20	88	53	0	14	N
	21	92	55	0	6	SE
	22	88	48	0	2	E
	23	71	43	0	2	SE
	24	78	48	0	4	S
	25	78	51	0	4	S
	26	75	56	0	4	S
	27	64	57	.01	5	S
	28	74	47	.10	5	W
	29	72	44	0	3	S
	30	68	43	.16	4	NE
Ave. Temp.	82.7	50.6		Ave. Wind Velocity	5.9	