

THE EFFECTS OF LOW LEVELS OF 2,4-D ON SUNFLOWER
AND RAGWEED SEED PRODUCTION IN NORTHEASTERN KANSAS

by

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INTRODUCTION

Farming practices were generally beneficial to game species prior to World War II. Fields and farms were small and interspersed among woodlands. This resulted in a habitat diversity where many game species thrived. Weeds were common in cropland and disturbed areas. Bobwhite (Colinus virginianus) was one species that did well in this type of habitat. Their populations declined after the 1940's (Rosene 1969) when fields became larger and diversity declined. The use of selective herbicides also began shortly after World War II, reducing weed numbers. Many of these plants produce seeds which are utilized by bobwhite.

Two weeds that are very important to the bobwhite in Kansas, common sunflower (Helianthus annuus) and ragweed (Ambrosia artemisiifolia), are very susceptible to 2,4-dichlorophenoxyacetic acid (2,4-D), one of the most-used herbicides in the United States. Ragweed is an especially unpopular weed due to its hayfever-producing pollen. Despite ragweed's value as an important wildlife food (Martin et al. 1951), Sylwester (1951:20) has stated "Any step we can take in the elimination of such unwanted plants as...ragweed should be encouraged."

While weed control is undoubtedly necessary in agricultural land, drift from herbicide spray application can be a problem (Nordby and Skuterud 1974). Drift can affect not only adjacent susceptible crops, but also seed producing weeds in nearby wildlife lands. Low levels of a synthetic auxin herbicide such as 2,4-D may increase or decrease plant growth depending on the level of herbicide used and the conditions under which it was applied. It is the purpose of this study to simulate drift on sunflowers and ragweed, and to determine the subsequent effect on seed production and quality.

The fruits of sunflower and ragweed are technically cypselas, but for simplicity will be called seeds in this report.

The literature on 2,4-D is voluminous. The more pertinent literature on each topic is reviewed in the following section.

LITERATURE REVIEW

Much of the early work in developing synthetic growth-regulating hormones (including 2,4-D) as herbicides was conducted by the United States Army during World War II. The potential for biological warfare (the destruction of enemy crops) was the basis for this research. The war ended before these herbicides could be so used, but this research facilitated the early post-war introduction of phenoxy herbicides for agricultural purposes (House et al. 1967).

Over 43.5 million pounds (19.7 million kilograms) acid equivalent of 2,4-D were produced in the United States in 1970 (Fowler and Mahan 1975), and in Kansas, 2,4-D is the most-dispersed herbicide by the State Weed and Pesticide Division (Kansas Department of Agriculture 1975:mimeo). This herbicide is used to control broadleaf weeds on range lands and in tolerant crops. The rates used generally vary between 1/2 and 2 pounds acid equivalent per acre (0.56 and 2.24 kg/ha) for barley, corn, oats, pastures, range land, sorghum, and wheat (Klingman et al. 1975:215).

Mode of Action

Robertson and Kirkwood (1969, 1970) review the mode of action of phenoxy-acid herbicides. After entering the leaf through either the cuticle or stomata, they indicate that the herbicide probably moves through the xylem within the treated leaf. It is then transported throughout the plant via the phloem system. Although 2,4-D affects plant respiration (inhibits oxygen uptake and ATP synthesis), photosynthesis (reduction of CO₂ absorption and inhibition of the Hill reaction), and mineral uptake, the probable primary mechanism of 2,4-D action is the increased synthesis of RNA and protein. This causes the often-seen stem and leaf epinasty and swelling of stems. Holm and Abeles (1968) have suggested that ethylene production in

response to 2,4-D treatment may be the cause of stem swelling and nucleic acid synthesis.

Growth is stimulated by low concentrations of 2,4-D and inhibited by high concentrations. Hanson and Slife (1969) argue that 2,4-D acts as an auxin but accumulates to higher concentrations than indoleacetic acid (IAA) because of its slow degradation. Either growth or inhibition of growth can occur depending on the tissues and concentrations of 2,4-D involved (Wort 1964).

Factors Affecting Action of 2,4-D

Humidity, light, and temperature are important in the absorption of herbicides. Pallas (1958) found that significantly more 2,4-D was absorbed and translocated in red kidney beans (Phaseolus vulgaris) at high humidities (70-74 percent) than at low humidities (34-48 percent) within a temperature range of 20-35°C. Clor et al. (1962) found increased translocation at high humidity using cotton (Gossypium barbadense) as a test plant. Water stress in bean plants did not greatly affect absorption of 2,4-D, but translocation was drastically reduced (Basler et al. 1961). They speculated that since stem elongation ceases under moisture stress, endogenous auxins are likely not translocated either. An actively growing plant is necessary for good translocation of an herbicide.

Numerous studies have been conducted concerning the increased activity of 2,4-D with increased temperature. For example, Muzik (1965) showed that tomato (Lycopersicon esculentum) leaves dipped in 2,4-D and kept at 70 and 80°F (21.1 and 26.6°C) exhibited typical 2,4-D responses (epinasty of leaves, twisting and swelling of stems, and tumorous outgrowths). Plants kept at 55°F (12.8°C) showed no visible response to the herbicide; growth continued at the same slow rate as controls. However, symptoms did

show up if the plants were transferred to 70-90°F (21.1-32.2°C) as long as two months after spraying. Pallas (1958) demonstrated an increase in absorption and translocation of 2,4-D with increasing temperatures between 20 and 30°C using red kidney beans. Barrier and Loomis (1957) increased absorption of 2,4-D in soybeans (Glycine max) by increasing temperature. This effect was also found by Marth and Davis (1945) in early winter cress (Barbarea vernea) and narrow-leafed plantain (Plantago lanceolata), and in red kidney beans, perennial rye, and crabgrass (Digitaria sp.) by Kelly (1949).

Light intensity affects the growth rate of plants and therefore is also important in the absorption and translocation of 2,4-D. Generally, plants kept under low light conditions are more susceptible to the herbicide than plants kept under high light conditions. Blackman and Robertson-Cunningham (1955) found that the LD₅₀ for plants was reduced by a factor of 10 under conditions of 12 percent of normal daylight intensity. Mitchell and Brown (1946) found similar results in bean plants and speculated that the translocation of 2,4-D was closely related to the translocation of organic food materials. Water hyacinth (Eichhornia crassipes) plants treated with 2,4-D also showed greater epinasty and necrosis in shaded plants (Penfound and Minyard 1947), although red kidney beans showed herbicide effects to a lesser extent. Etiolated bean plants did not translocate 2,4-D unless sugar in solution was applied to the leaves (Rohrbaugh and Rice 1949), further substantiating Mitchell and Brown's (1946) results.

The formulation of the herbicide and any surfactants used have an obvious effect on the action of that herbicide. Amine salts and esters are the most common formulations of 2,4-D used (Klingman et al. 1975:216). Although esters are generally more efficiently absorbed by leaves than amines (Robertson and Kirkwood 1969), problems with volatility may be encountered

when esters are used near susceptible non-target plants (Klingman et al. 1975, QueHee and Sutherland 1975). Amines pose virtually no volatility problems (Klingman et al. 1975). Other formulations used to a much lesser degree include emulsifiable acid and mineral salts. Surface active agents (surfactants) are used in conjunction with herbicides for many purposes (sticking, wetting, detergency, etc.), but they are ultimately used to increase the absorption of foliar applied herbicides.

Many authors have discussed the breakdown and persistence of 2,4-D. Bell (1956) and Crosby and Tutass (1968) discuss the rapid decomposition of 2,4-D under ultraviolet light. Klingman et al. (1975:76) list the persistence of biological activity as one month or less for 2,4-D. Lavy et al. (1973) found that essentially all the 2,4-D applied at various depths in the soil had dissipated within five months in Nebraska, while Wilson and Cheng (1976) found small quantities (4 percent) remaining in Washington soils 6 months after spraying. Wilson and Cheng's results were from a very dry year, and 2,4-D persists longer in dry soil (Brown and Mitchell 1948). Brown and Mitchell also found increasing temperatures and increasing amounts of organic materials in the soil to decrease the persistence of 2,4-D in the soil. Microorganisms assist in the breakdown (Newman et al. 1952) and this was shown by the longer persistence of 2,4-D in autoclaved soil (Brown and Mitchell 1948). Morton et al. (1967) have shown that 2,4-D also has a very short half-life in green tissue (two weeks in silver beardgrass (Andropogon saccharoides), little bluestem (Schizachyrium scoparius) and dallisgrass (Paspalum dilatatum)), and virtually disappears after 16 weeks.

The ultimate effect of an herbicide on plant growth is also dependent on the size of the spray droplets. In recent studies, it has been shown that small herbicide spray-droplets have a greater effect on plants than

large spray-droplet sizes. Small (< 0.1 mm diameter) spray droplets of 2,4-D reduce soybean seed production significantly more than large (< 0.3 mm diameter) spray droplets (Ennis and Williamson 1963). Using an insecticide (Malathion), Polles and Vinson (1969) found increased penetration of the leaf cuticle with smaller droplet sizes and speculated that it was due to the greater insecticide surface area involved for leaf absorption when constant volumes of spray were used. Way (1969) found that small droplets (100 microns) had a greater visible effect on lettuce (*Lactuca serriola*) using 2-methyl-4-chlorophenoxyacetic acid (MCPA) than large droplets (500 microns). Sunflower also showed greater epinasty when sprayed with small droplet sizes of 2,4-D (100 microns) than with large droplet sizes (200 and 400 microns) (McKinlay et al. 1972). They speculated that larger droplets killed the leaf cells directly under them, thus reducing translocation. The total leaf area contacted by a given volume of herbicide would also be greater with smaller droplets. In the study by McKinlay et al., six times as much active ingredient was needed for a given effect using 400 micron droplets as compared to 100 micron droplets.

Increased volatility of small droplets of esters of 2,4-D (QueHee and Sutherland 1975) and drift of small spray droplets are two problems of spraying with small droplet sizes. Volatility can reduce the intended effect of the spray and/or cause damage to adjacent desirable flora. Small spray particles drift a greater distance than large particles and can also cause damage to adjacent flora. Reimer et al. (1966) estimated that a 300 micron droplet falling 50 feet (15.2 m) with a 5 mph (8.1 kph) wind would drift about 100 feet (30.5 m). Brooks (1947) estimated that a 100 micron droplet falling 10 feet (3.1 m) with a 3 mph (4.8 kph) wind would drift 409 feet (124.7 m). Nordby and Skuterud (1974) state that under good

conditions, spray drift of up to 6 percent of the volume can be expected, and half of this would be within a strip 10 m from the end of the tractor-pulled boom. Under most spray conditions, they estimate drift to amount to 1.4-37 percent of the spray volume applied. With drift this prevalent, and the effects of smaller droplet sizes significantly increasing the effect of spray, drift can have an effect on adjacent vegetation and therefore on wildlife.

Effects of 2,4-D on Wildlife

Spray drift can affect wildlife either directly or indirectly. Since the publication of Silent Spring (Carson 1962), direct effects of pesticides on wildlife (not just through spray drift) have been widely publicized. Direct effects might come about due to an animal ingesting or coming in contact with a pesticide. In his article assessing the hazards of chemical brush control, Norris (1971:720) concludes "I am satisfied after evaluating the toxicity of 2,4-D, amitrole, 2,4,5-T and picloram that the proper use of these herbicides will not normally result in either an acute or chronic hazard to nontarget organisms on forest lands." At normally applied rates, then, 2,4-D appears relatively safe.

Acute oral toxicity (LD_{50} ; single dose which is lethal to 50 percent of the animals tested) of the various formulations of 2,4-D range from 300 to 1000 mg/kg (Klingman et al. 1975). Tucker and Crabtree (1970) found LD_{50} values of 472 mg/kg for pheasant (Phasianus colchicus), 668 mg/kg for coturnix (Coturnix coturnix) and pigeon (Columba livia), and 1000 mg/kg for mallard (Anas platyrhynchos) for 2,4-D technical acid. Mallards given the amine base showed an LD_{50} of about 2000 mg/kg. Heath et al. (1972) lists the LC_{50} (lethal concentration in a 5-day diet to kill 50 percent of the animals tested) for bobwhite of all formulations of 2,4-D as greater than

5000 ppm. Another study with bobwhite found an LC_{50} of 38,000 mg/kg for adult birds and 28,000 mg/kg for young birds (Fish and Wildlife Service 1960). These levels are considerably above those that would be normally encountered by organisms in the wild.

Eggshell thinning has been shown to occur following oral doses of 2,4-D (Haegele and Tucker 1974). Coturnix given a single oral dose of 250 mg/kg produced eggshells which were 50 microns thinner than the normal 200 micron-thick shells of control birds for two days following treatment. Likewise, mallards given 1500 mg/kg produced eggshells that were 30 microns thinner than the normal 330 micron thickness for two days following treatment. Eggshells produced more than two days after treatment were normal in thickness. Chickens given 150 mg of 2,4-D/kg of food showed no adverse effect on egg production, egg or yolk weight, eggshell thickness, or hatchability or growth rate of progeny (Whitehead and Pettigrew 1972). They concluded (concerning 2,4-D) saying "...at the still lower dose rates more likely to be encountered in normal agricultural practice, significant impairment of production is unlikely to occur."

One reason for the short term effect of 2,4-D found by Haegele and Tucker (1974) is the rapid elimination of 2,4-D by animals. Levey and Lewis (1947) found that 60 percent of the phenoxyacetic acid fed to rabbits showed up in excreta within 6 hours of feeding, and 96 percent of it was excreted in 24 hours. Similar results were found with rats (Khanna and Fang 1966) and mice (Zielinski and Fishbein 1967). In sheep (Clark et al. 1964), over 97 percent of an orally administered dose (4 mg/kg) was excreted unchanged in 72 hours.

Recently, teratogenic effects have been found in 2,4,5-T-fed mice. Significant increases of cystic kidney and cleft palate were observed when

mice in day 6 of pregnancy were given 113 mg/kg of 2,4,5-T (Courtney et al. 1970). However, 2,4,5-T did not produce teratogenic effects on chick embryos (Strange and Kerr 1976). Embryotoxicity did occur in rats using 2,4-D, but teratogenic effects did not appear even at levels approaching the LD₅₀ for rats (Schwetz et al. 1971). Khera et al. (1971) indicated that 2,4-D may cause teratogenic effects (skeletal malformations) in white Wistar rats at high dosage levels. These results, however, were preliminary. In summary, 2,4-D poses little direct hazard to wildlife.

Effects of 2,4-D on Plants

Herbicides may influence wildlife indirectly, through habitat change. Effects may differ in differing areas. Martin (1965) found a slight increase in bobwhite populations after a post oak-black oak woodland in Oklahoma was sprayed with 2,4-D and 2,4,5-T. High populations of native grasses and forbs (Ambrosia psilostachya was common) followed the spraying. Forbs and grasses invaded the understory opened up by the dying oaks. When range land consisting of forbs and grasses is sprayed with 2,4-D, forbs are generally eliminated. This has been shown to cause declines in food for pocket gophers (Thomomys talpoides), causing population reductions of approximately 90 percent (Keith et al. 1959, Hull 1971). Johnson and Hansen (1969) found deer mouse (Peromyscus maniculatus) populations were affected very little by 2,4-D treatment of range lands in Colorado. Species dependent upon forbs decreased while species dependent on grasses increased: pocket gopher and least chipmunk (Eutamias minimus) populations declined while montane vole (Microtus montanus) populations increased.

Use of 2,4-D for brush control for livestock can affect many species. Spraying in large blocks has been shown to reduce sage grouse (Centrocercus urophasianus), while strip spraying of sagebrush did not affect use of an

area by adults (Carr and Glover 1970). Young grouse (chicks) were not found in any sprayed areas. Southwood and Cross (1968) found a significant decrease of arthropod numbers and species in sprayed areas in England. Arthropod numbers decreased by more than $2/3$, causing a reduction in juvenile survival in partridge (Perdix perdix). MCPA and 2,4-D were the herbicides used. Brewers sparrow (Spizella breweri), dependent on sagebrush for nesting cover, decreased with sagebrush control (Best 1972). He also speculated that the decrease in floral diversity due to 2,4-D would probably cause a reduction in the number and/or species of insects present which Brewers sparrows utilize at certain times of the year.

Most of the native species important as bobwhite food in Kansas listed by Robel (1963) are weeds associated with agriculture and disturbed areas. Nearly all of these are very susceptible to 2,4-D (U.S. Department of Agriculture 1974). Bobwhite, as well as many other wildlife species, utilize these plants in agricultural areas and in areas adjacent to agriculture. Herbicide drift into adjacent areas can occur when spraying weeds in agricultural areas, affecting quail food sources. Very little work has been done on this problem.

Sunflower and ragweed, whose seeds are important food sources for bobwhite in Kansas, are very susceptible to 2,4-D. Throughout much of the bobwhite's range, seeds of common ragweed are one of the most consumed fall and early winter foods (Martin et al. 1951:421). Robel (1963) found ragweed to be the second most important fall (November and December) food in Kansas. By volume, ragweed was the most utilized seed in Virginia (Baldwin and Handley 1946) and Tennessee (Cody 1944), second in Kentucky (Barbour 1951) and Nebraska (Damon 1949), third in Missouri (Korschgen 1948), seventh in the southeastern United States (Davidson 1942), and very important in Oklahoma

(Bird and Bird 1931, Baumgartner et al. 1952), Ohio (Schultz 1949), and Illinois (Barnes and Klimstra 1964, Ellis et al. 1969). In addition, ragweed was highest in frequency of occurrence and volume in disturbed areas in Georgia (Brunswig and Johnson 1973). Other species of ragweed (A. bidentata, A. trifida, and A. psilostachya) can also be locally important food sources to quail (Korschgen 1948, Baumgartner et al. 1952, Wagner 1949, Lehmann and Ward 1941). The effects of low levels of 2,4-D (such as those due to drift) on ragweed seed production are not known.

Where the ranges of sunflower and bobwhite overlap, sunflower seeds are also an important food source. Sunflower seed was found important in Kansas (Robel 1963), Nebraska (Damon 1949), and Oklahoma (Bird and Bird 1931, Baumgartner et al. 1952). Sunflower can also be locally important in Texas (Lehman and Ward 1941) and Missouri (Korschgen 1948). The effect of drift-like levels of 2,4-D on subsequent seed production of young sunflower plants is unknown.

There have been numerous studies concerning the effect of low levels of 2,4-D on plant growth. Weaver (1946) concluded that 2,4-D delayed the onset and decreased the amount of seed production in red kidney beans and soybeans. He also found that low levels of 2,4-D increased the diameters of hypocotyls and first internodes, increased the weight of primary leaves, and decreased the weight of trifoliate leaves. In a similar study, Weaver et al. (1946) found significant reductions in soybean seed production when the plants were treated in the flowering or early pod stage with 0.5 lb/acre (0.56 kg/ha) of 2,4-D. Spraying when plants were 6-8 inches high did not significantly reduce yield, however younger plants showed greater visible response (epinasty, etc.) than plants treated at a later stage. Baker (1969) found similar results using DSMA and MSMA on cotton.

Slife (1956) found that young (2-4 leaf stage) ragweed plants were more susceptible to 2,4-D than older plants. A rate of 1/8 lb/acre (0.28 kg/ha) of 2,4-D did not control subsequent forb (predominantly ragweed) seed production in Michigan corn fields, while 1/3 and 1/2 lb/acre (0.37 and 0.56 kg/ha) were effective (Jameson and Wheeler 1951). They did not state the stage of growth of the ragweed, but corn was 6-12 inches high at the time of spraying. Peters and Lowance (1970) showed that a decrease in ragweed seed production occurred when plants were sprayed with 0.56 or 1.12 kg of 2,4-D amine/acre. Control was enhanced when younger ragweed plants (15-20 cm) were sprayed. Soybeans sprayed with sublethal (0.015 and 0.150 kg/ha) levels showed non-significant differences from controls when seed yields (kg/ha) were compared (Basnet et al. 1972). Treated plants produced fewer pods, but this was offset by an increased weight/100 seeds ($P < 0.05$). Plants were sprayed at one-tenth bloom stage.

Many authors have studied subsequent germination of seeds from treated plants, and it seems that germination is only affected when plants are sprayed at the seed-filling stage. For example, Rojas-Garciduenas and Kommendahl (1960) found that pigweed (Amaranthus sp.) plants sprayed with 2,4-D at the time of flowering produced seeds that germinated more rapidly than non-treated seeds. Evans et al. (1963) found that amitrole, DMPA, and dalapon seriously reduced seed production and subsequent germination in medusahead (Elymus caput-medusae) when applied at flowering time. Germination, seed production, and weight per seed were significantly reduced when curly dock (Rumex crispus) was sprayed at anthesis (Maun and Cavers 1969). Herbicides are translocated into the seed when photosynthate is being transported into the seed, but the herbicides are not mobile when the "sink" (seed) develops if they are applied before flowering (Thompson and Egli 1973).

Male sterile flowers have also been shown to be induced by 2,4-D when applied before and during flowering (Rehm 1952). Smith et al. (1946) and Grigsby (1945), using 2,4-D, prevented pollen shedding in ragweed. The levels of 2,4-D used, however, were very nearly lethal to the plants. Grigsby (1946) severely reduced seed production in ragweed by spraying with 250 ppm of 2,4-D at the stage just preceeding flower stalk elongation. He found 500 ppm was sufficient to kill young plants and to stop pollen production in older plants.

Several authors have considered the problem of spray drift and subsequent seed production in susceptible plants. Greenshields and White (1954) and Greenshields and Putt (1958) simulated spray drift on sweet clover (Trifolium sp.) and cultivated sunflower, respectively, using both amine and ester formulations. These plants were kept downwind of a 2,4-D spray operation. Seed production was delayed and seed quality was reduced in sweet clover when exposed to 2,4-D drift just prior to flowering. Cultivated sunflowers planted 12 May and exposed to drift on 26 June showed reduction of seed production (19 percent) even at distances of 120 rods (603.5 m) from the spraying. Wind velocity was 9-12 mph (14.5-19.3 kph) and a spray pressure of 30 psi was used. Reduction in seed yield was due to small head size, rows of empty seeds in the heads, and lighter weight of seeds. Betts and Ashford (1976) found that while 2,4-D did not reduce subsequent germination of turnip rape seeds from plants exposed to drift before flowering time, seed yield was significantly reduced. No work of this kind has been done with native sunflowers or with very low levels of 2,4-D on ragweed. Very little work has been done on the quality of seeds (protein content, caloric content, etc.) produced by plants subjected to spray treatment.

METHODS AND MATERIALS

The Study Area

Sunflower and ragweed have been shown to be very important to bobwhite in Kansas and were therefore the species chosen to study the effects of low levels of 2,4-D on subsequent seed production. High densities of sunflower and ragweed were needed for the study, and these were located on the north-eastern corner of the Fort Riley Military Reservation, 11 km southwest of Manhattan, Kansas. The native vegetation of this area of the western edge of the Flint Hills is prairie. Johnson and Robel (1968) list important plant associations. Although some cultivated crops are grown in the Flint Hills, the primary agricultural practice is grazing livestock.

Because large numbers of sunflower and ragweed plants could not be found in the same area, two study areas were chosen: one with high densities of sunflower and one with high densities of ragweed. The sunflower area was approximately 3 1/2 km north-north-west of the Manhattan Municipal Airport USGS Bench Mark. The area was 150 x 9-m in size and was bounded 20 m on the south by a wind break of primarily Osage Orange (Maclura pomifera) and hackberry (Celtis occidentalis), on the east by a cultivated field of sorghum, and on the north and west by an open field characterized by grasses and other herbaceous cover.

This area was part of a sorghum food-plot program for wildlife carried on by Fort Riley personnel. Sunflowers invaded when the sorghum plot was discontinued. Controlled fires frequently burned the area to minimize woody vegetation. Controlled fires burned the area in mid-March 1976 and in spring 1973.

Soil in the sunflower area is Wymore silty clay loam, with 0-1 percent slopes. This soil is characterized by a high water-holding capacity with a

fine textured subsoil which is slowly permeable. Water commonly ponds after heavy rains. This soil may also restrict the growth of roots (U.S. Dept. Agriculture SCS 1975).

The second area chosen for study was the center of a large area densely populated with ragweed. This area was approximately 2.8 km north of the sunflower area, and was 30-m E-W x 50-m N-S in size.

Fire and the control of wild fires are important in this area. It has been irregularly burned in the past and burned in March 1976 during this study. The area has been plowed annually in the spring (April 1976 during this study) to keep vegetation at a minimum for a fire break.

The rolling topography of this area is in contrast to the flat sunflower area. Slope on the study area is approximately 5 percent and south-facing. Soils are of the Benfield-Florence complex, characterized by low soil water capacity. These soils readily absorb water and release it for plant utilization (U.S. Dept. Agriculture SCS 1975). Limestone fragments occur where hillside plowing has caused erosion.

Procedures

Both areas were divided into 30 plots. In 1975, the sunflower plots measured 4.5 x 9-m and the ragweed plots measured 5 x 10-m. To facilitate spraying in 1976, plots were reduced to 2 x 3-m.

During summer and fall 1975, seed production data were collected from both sunflower and ragweed areas. Twelve plants were randomly selected in each plot, making a total of 360 plants of each species.

A material similar to bridal veil (12.5 holes/cm) was used to cover ragweed plants and sunflower seed heads in order to prevent avian predation and loss of seed. Entire ragweed plants were covered shortly after flowering took place (the week of 10 September) and collected during early November.

All flower heads on the selected sunflower plants were also wrapped or collected shortly after each flowered, and then separated into five different groups on the basis of flowering time: 18 August, 1 September, 23 September, and 15 November. The bagging on 23 September was divided into two parts. Fruiting had already taken place in many of the flower heads, and these were collected and labeled 23 September (unbagged). The others were bagged and left in the field until the middle of November (labeled 23 September (bagged)). All seed heads were collected and brought to the lab by 15 November. In addition, all seeds were collected from 30 randomly selected sunflower plants which were not bagged to determine the effect of this covering material on seed production.

Seeds were separated from other plant material, insect-predated seeds were separated from intact seeds, and both were counted. Air dry weight of seeds was then measured to 0.1 mg on a per plant and a per flower head basis for ragweed and sunflower, respectively. In addition, diameters of sunflower seed heads were taken with a calipers to the nearest 0.1 mm. Due to the ovoid shape of the capitula, two diameters (the longest diameter and one perpendicular to it) were taken.

Sunflower and ragweed populations were estimated in their respective plots. All flowering ragweed plants were counted in $10-0.1 \text{ m}^2$ subplots systematically located in each of the 30 ragweed plots in 1975 (6 population estimates were taken per plot in 1976). All flowering sunflower plants were counted in $8-1 \text{ m}^2$ subplots systematically located in each of the 30 sunflower plots in 1975 (4 population estimates/plot in 1976). Plant height, number of flowers, and flower stems broken due to predation were also recorded for sunflower in 1975.

Rainfall and soil moisture data were collected throughout the summers of 1975-76. One rain gauge was situated in each study area and both were checked within 12 hours after each rain. Due to rocky soil in the ragweed area, soil moisture was taken only in the sunflower study area. Soil samples were taken weekly (biweekly in 1975) at depths of 10-20, 30-40, and 50-60 cm. Soil moisture was determined using the gravimetric method (Buckman and Brady 1969:177).

In 1976 several changes in procedure were made. Covering material was not used to protect seeds. Plots were visited two or three times weekly and ripe sunflower seed heads were collected to prevent loss of seed. In addition, 17 plants were randomly selected in the sunflower area plots 1-21, and 15 in plots 22-30 (these latter plots had low plant densities). Twenty-five plants were randomly selected per plot in the ragweed area.

On 31 May and 1 June 1976 plants were measured before spraying in the sunflower and ragweed areas, respectively. Plant heights and numbers of pairs of leaves were measured for a systematically selected group of plants in both areas. All plants were measured in 15-1-meter square areas for sunflower and 12-0.1-meter square areas for ragweed. These plants were remeasured on 15 June to determine the immediate effects of the spray on plant growth.

An amine salt formulation of 2,4-D (Dow Formula 40 Herbicide: 4 lbs acid equivalent per gallon) was used to prevent volatility problems. Two Hudson Stainless Steel Climax 6335S sprayers (H. D. Hudson Manufacturing Company, Chicago, Illinois) were used at approximately 18 pounds pressure.

A spray level was desired that would be similar to levels incurred by plants due to spray drift and a level which would have some effect on the plants without killing them. The levels of 2,4-D selected that would

fit these criteria were 50 and 100 ppm of 2,4-D sprayed at 0.1 liters/m² (approximately 0.050 and 0.100 kg acid equivalent per hectare or 1/10 to 1/5 of the recommended application rate).

Plots were grouped into blocks of three plots each. Within each block, the three treatments were randomly assigned. A 1-meter wide area around the outside of treated plots also received treatment to reduce any possible edge effect. There were 10 blocks in both sunflower and ragweed areas.

Spraying of sunflowers began at 6:30 a.m. 1 June. Temperature was 16°C at spray time, and winds were calm. Humidity was 100 percent, and there was moderate dew on the ground. Skies were mostly sunny all day. Soil moistures in the sunflower study area were 27.11, 32.77, and 28.75 percent for 10-20, 30-40, and 50-60 cm soil depths, respectively. Mean height of plants was 21.7 cm and most plants had 2 to 4 pairs of leaves. Ragweeds were sprayed 2 June beginning at 6:15 a.m. Temperature was 18°C at spray time. Wind, humidity, dew, and light conditions were similar to 1 June. Mean height of ragweed plants was 11.9 cm and most ragweed plants had 2 to 4 pairs of leaves. High temperatures were 29°C on 1 June and 28°C on 2 June. Weather data for the week following spraying are given in Appendix 3.

Protein, moisture, ash, fat, fiber, and digestable carbohydrates in sunflower and ragweed seeds were determined using a proximate analysis. In 1976, seeds from plants under each treatment regime were tested to determine the effects of 2,4-D on seed composition.

Energy content of seeds was determined using bomb calorimetry. Whole seeds were ground to a consistent particle size and 1-gram samples were combusted in a Parr 1200 calorimeter under 30 atmospheres of oxygen. In 1975, energy values for sunflower were determined separately for each time

period of flowering following methods described by Johnson and Robel (1968). In 1976, determinations of seed energy content were conducted on seed samples collected from each treatment.

While counting sunflower seeds it was noticed that seeds from treated plants appeared darker than seeds from untreated plants. Seeds produced later in the year also appeared darker. A color chart (Munsell Book of Color) was obtained and a series of colors resembling seed color (2.5 Y Hue: yellow-gray) were numbered from lightest to darkest. Seeds were taken from each treatment and time and matched to the color chart. Means were then calculated for each treatment and time. Replications from plants and flowers could not be done because color differences were not noticed until after all seeds were put in containers with respect to their treatment and seed-ripening time. Two-dimensional paper chromatography on treated and untreated seeds was conducted to determine if color differences could be explained by differences in seed phenolic compounds. The method used (Mabry et al. 1970) detects most phenolic compounds and is a qualitative rather than a quantitative method.

Two-way analysis of variance was used to test treatment effects on the various production parameters. Treatment sums of squares were partitioned into linear and quadratic (nonlinear) components (Snedecor and Cochran 1967), and F-tests were performed on each. The probability of a greater F-value for the linear component will be denoted as P_L and for the nonlinear component as P_{NL} . Since plots were the replicates in this experiment and not plants or flowers, the interaction mean square was used as the error term for calculating F-values.

RESULTS: 1975

Sunflower Seed Production

All seed heads were collected from 360 randomly selected sunflower plants in 1975. Number of seeds per capitulum ranged from 0 to 344 with a mean of 26.18 (standard error = 0.89, Table 1). Capitula averaged 3.15 seeds that were partially consumed by insects. Robel and Linderman (1966) determined that bobwhite consume primarily intact seeds, therefore the number of seeds potentially available for bobwhite was 23.03 per flower head. Weight of these intact seeds per flower head was 0.126 grams, and the weight per seed, calculated by division, was 5.47 mg.

Production in the sunflower area was calculated by multiplying the number of flower heads/m² (35.78) by the weight of seed/flower head (0.126) and converting to kg of seed/100 m². In 1975, sunflower production was 0.45 kg of seed/100 m².

A comparison was made between the time of flowering and production for sunflower (Table 2). Production was highest in the 1 September and 23 September (bagged) periods. Average number of seeds produced in the 1 September and 23 September (bagged) periods was 46.07 and 42.32 seeds/flower head, while the other 3 periods produced less than 10 seeds/capitulum each. An LSD test showed the 18 August, 23 September (unbagged), and 15 November periods had significantly lower seed production per flower head ($P < 0.05$) than the 1 September and 23 September (bagged) periods.

A comparison was made between bagged and unbagged sunflowers to determine the effect of this covering. Many seeds were lost due to not bagging, but it was possible to estimate this number by examining the seed head. If 90 percent of the missing seeds were counted (allowing for predation), virtually no difference was found in the number of unpredated

Table 1. Sunflower and ragweed seed production data collected on the Fort Riley Military Reservation in 1975.

<u>Sunflower</u>	<u>Number of Capitula</u>	<u>Mean $\pm$ Standard Error</u>
Total seeds/capitulum	1957	26.18 $\pm$ 0.98
Predated seeds/capitulum	1957	3.15
Unpredated seeds/capitulum	1957	23.03 $\pm$ 0.89
Weight of seeds/capitulum (g)	1957	0.126 $\pm$ 0.029
Weight/seed (mg)	--	5.47
Population (capitula/m ²)	236	35.78 $\pm$ 1.79
Production (kg of seed/100 m ²)	--	0.45
<u>Ragweed</u>	<u>Number of Capitula</u>	<u>Mean $\pm$ Standard Error</u>
Total seeds/plant	351	36.58 $\pm$ 2.66
Predated seeds/plant	351	1.28
Unpredated seeds/plant	351	35.29 $\pm$ 2.63
Weight of seeds/plant (g)	351	0.113 $\pm$ 0.009
Weight/seed (mg)	--	3.15
Population (plants/0.1 m ²)	300	25.91 $\pm$ 1.18
Production (kg of seed/100 m ²)	--	2.93

Table 2. Sunflower seed production by flowering time in 1975. Flowering times are dates when flower heads were wrapped to prevent loss of seed: 18 August, 1 September, and 23 September (bagged), and when ripe flower heads were collected without wrapping: 23 September (unbagged) and 15 November.

Flowering time	N	Seeds per flower head (mean $\pm$ std. error)	Predated seeds per flower head	Weight per seed (mg)
18 August	410	6.37 $\pm$ 0.73	0.63	5.7
1 September	245	46.07 $\pm$ 3.73	5.45	5.9
23 September (unbagged)	466	8.13 $\pm$ 0.95	0.72	4.2
23 September (bagged)	787	42.32 $\pm$ 1.76	5.33	5.5
15 November	49	3.76 $\pm$ 1.80	0.39	3.1

seeds produced per flower head (23.03 and 24.36 for bagged and unbagged, respectively). Calorimetry did show a difference (5805.45 cal/g for unbagged and 5915.48 cal/g for bagged), but since only enough seeds were available for two energy determinations, significance was not tested. This difference may have been due to the larger seeds near the center of the seed head falling to the ground. Large seeds probably have more energy/g than small seeds because the surface area/volume ratio becomes smaller as seed size increases and most of the fats and oils in a seed are stored in the seed endosperm.

Ragweed Seed Production

The mean number of seeds produced by the 351 ragweed plants collected in 1975 was 36.58 (standard error = 2.66, Table 1). Ragweed plants were collected only if they produced seed, and the range in seed production per plant was 1 to 574. An average of 1.28 seeds/plant were partially consumed by insects, leaving 35.29 undamaged seeds per plant. The average weight of ragweed seeds was 0.113 g/plant, and the weight per seed was then calculated as 3.15 mg.

Production in the ragweed area was calculated by multiplying the number of seed bearing plants/ 0.1 m^2 (25.91) by the weight of seed per plant (0.113) and then converting to kg of seed/ 100 m^2 . In 1975, ragweed production was 2.93 kg of seed/ 100 m^2 .

Predation

Two types of insect predation occurred on the sunflowers. In one, the insect laid its eggs in the seeds and the larvae consumed the inside of the seed prior to emerging. This type of predation also occurred in ragweed. The second type of predation was the girdling and subsequent breakage of a flower stem by insects. Although this type of predation was not observed, the insect involved was probably a snout beetle (Coleoptera: Curculionidae).

Seed predation accounted for 3.15 seeds per capitulum in sunflower (12.0 percent of the total seeds produced) and 1.28 predated seeds per plant in ragweed (3.5 percent of the total seeds produced). Before some larvae emerged, frost or another mortality factor undoubtedly occurred. Since predated seeds were counted by observing holes in the seeds, the predation estimate is probably somewhat conservative. Seeds entirely consumed by insects, although probably few in number, would also tend to make the predation estimate conservative.

Flower predation of sunflower caused the loss of an average of 0.90 flower heads per plant, or 18.1 percent of the total flowers produced. Assuming the beetles selected flower stems at random, the two types of predation combined to reduce sunflower seed production 27.9 percent.

Seed Composition

Proximate analysis showed that ash, fat, and fiber content in sunflower and ragweed were very similar (Table 3). Protein content of ragweed was higher than sunflower (23.0 and 17.7 percent, respectively), while digestible carbohydrate content was higher in sunflower (23.0 and 17.1 percent, respectively).

Energy values obtained through bomb calorimetry were slightly higher for sunflower seed (5915.48 cal/g) than for ragweed seed (5843.96 cal/g) (Table 4). One-way analysis of variance showed significant ($P < 0.01$) differences between time of flowering and caloric content for sunflower. Those seeds produced during the 1 September and 23 September (bagged) periods for sunflower were considerably higher in caloric content (5997.37 and 5919.35 cal/g) than those produced in the 18 August and 23 September (unbagged) periods (5770.55 and 5571.79 cal/g). Not enough seeds were collected from the 15 November period to determine caloric content. Predated seeds contained an average of 5195.34 cal/g.

Table 3. Composition of sunflower and ragweed seeds collected in 1975. Percent composition was detected by proximate analysis.

Species	Percent Composition				
	Moisture	Protein	Ash	Fat	Fiber
Ragweed	3.2	23.0	3.2	21.0	32.5
Sunflower	2.3	17.7	3.4	21.2	32.4
					Digestible Carbohydrates
					17.1
					23.0

Table 4. Caloric content of sunflower seeds collected from 4 flowering times and caloric content of ragweed seeds in 1975.

<u>Species and Time</u>	<u>Mean cal/g $\pm$ Standard Error</u> ¹
<u>Sunflower</u>	
18 August	5770.55 $\pm$ 15.29 a
1 September	5997.30 $\pm$ 9.70 b
23 September (unbagged)	5571.79 $\pm$ 9.69 c
23 September (bagged)	5919.35 $\pm$ 5.54 d
Average	5915.48 $\pm$ 7.31
<u>Ragweed</u>	5843.96 $\pm$ 8.52

¹Means followed by different letters are significantly (P < 0.05) different.

RESULTS: 1976Ragweed Production

The effects of 2,4-D on ragweed plants were more severe than on sunflower plants. Leaves of ragweed plants were folded into the stem and stems were twisted on plants from both 2,4-D treatments. After a week, the plants sprayed with 50 ppm appeared almost completely recovered, and the plants sprayed with 100 ppm were showing signs of improvement. By 10 days following treatment, most treated plants showed no visible signs of 2,4-D damage (except for some twisted stems on plants treated with 100 ppm). No plants were observed to die due to treatment in the ragweed study area.

Plant growth in height was reduced following spray treatment. Between 1 June and 15 June, change in height averaged 7.52 cm for controls, 3.70 cm for 50 ppm, and 3.52 cm for the 100 ppm treatment. While not significant ($0.05 < P < 0.10$), it appears that both 2,4-D treatments reduced growth during this period.

Spray levels reduced plant height significantly by the end of the growing season ($P_{NL} < 0.01$). Average heights were 48.59 cm for control plants, while 2,4-D treated plants averaged 40.00 and 38.59 cm for 50 and 100 ppm, respectively, at the end of the growing season.

The mean number of seeds produced per plant in all ragweed plots was 81.43 in 1976. Two-way analysis of variance showed no significant difference ($P_{NL} = 0.3113$) in seed production between the three treatment levels (94.39 seeds/plant for controls and 68.93 and 81.52 seeds/plant for the 50 and 100 ppm treatments of 2,4-D, respectively, Table 5, 20).

Weight of seeds per plant did not vary significantly ($P_{NL} = 0.3769$) among treatments. Mean weights were 0.234 g/plant for controls, 0.176 g for plants treated with 50 ppm of 2,4-D, and 0.209 g/plant for the 100 ppm

Table 5. Ragweed production in 1976 under three 2,4-D treatment levels. Treatment sums of squares were partitioned into linear and non-linear, and probabilities of a greater F value are given for each.

	Treatment Means			Probabilities	
	Control	50 PPM	100 PPM	P _{lin}	P _{nonlin}
Final plant height (cm)	48.588	39.999	38.586	0.1003	0.0000
Branches > 12 cm length	2.050	1.742	1.990	0.8646	0.3613
Seeds/plant	94.389	68.929	81.523	0.5483	0.3113
Unpredated seeds/plant	92.387	67.619	79.980	0.5555	0.3144
Seed weight/plant (g)	0.234	0.176	0.209	0.6725	0.3769
Weight/seed (mg)	2.274	2.329	2.392	0.4669	0.9555
Predated seeds (%)	1.776	2.179	1.712	0.9450	0.5892

treatment. There were also no significant differences ($P_L = 0.4669$) between weight per 100 seeds. With increasing level of 2,4-D treatment, however, there was a slight increase in weight (0.2274 g/100 seeds for controls, and 0.2329 and 0.2392 g/100 seeds for the 50 and 100 ppm treatments, Table 5, 20).

The number of branches greater than 12 cm in length did not differ significantly between treatments ($P_{NL} = 0.3613$). Mean numbers of branches were 2.05 for controls and 1.74 and 1.99 for plants treated with 50 and 100 ppm of 2,4-D.

The predation rate (seeds predated/total seeds) for ragweed seeds in 1976 was 1.89 percent. No significant difference ($P_{NL} = 0.5892$) was found among treatments (1.78 percent predated for controls and 2.18 and 1.71 percent predated for 50 and 100 ppm of 2,4-D).

Total seed production was shown to be correlated with number of branches over 12 cm in length ($r = 0.6751$, $P < 0.01$). Seed production was also significantly correlated with plant height ($r = 0.5154$, $P < 0.001$). Plant height and number of branches were also positively correlated ($r = 0.4240$, $P < 0.01$) (Table 6).

Tall plants and plants which produced greater numbers of seeds also produced heavier seeds. Weight per 100 seeds was highly correlated ($P < 0.01$) with plant height ($r = 0.1892$), total number of seeds ($r = 0.2242$), and seed weight/plant ($r = 0.3395$) (Table 6). Number of branches were also positively correlated with weight per 100 seeds ($r = 0.0903$, $P < 0.05$).

Although seed weight fell off considerably in 1976 compared to 1975 (2.27 and 3.15 mg, respectively), overall weight of seeds per plant increased from 0.113 g to 0.234 g. This was due to the increased number of seeds produced per plant in 1976 (81.43) compared to 1975 (36.58). Populations

Table 6. Correlation coefficients of ragweed production parameters in 1976.

[illegible]

* - Significant at the 0.05 level.

** - Significant at the 0.01 level.

of ragweed were only 8.33 plants/0.1 m² in 1976, and production was 1.72 kg of seed per 100 m².

Seed composition:

Caloric content of ragweed seeds in 1976 differed ($P < 0.01$) between treatments, varying from 5651.20 cal/g in the controls to 5524.84 and 5618.20 cal/g in the 50 and 100 ppm treatments (Table 7).

Proximate analysis determined that moisture, ash, and fiber content were very similar between the three treatments, but protein content appeared to increase with treatment (22.1 percent for controls, and 25.8 and 25.1 percent for the 50 and 100 ppm treatments, respectively, Table 8). Fat content and digestible carbohydrate content appeared to decline with 2,4-D treatment. Fat content was 17.6 percent for controls, 16.0 percent for 50 ppm, and 16.4 percent for 100 ppm, while digestible carbohydrate levels were 20.1 percent for controls, 18.3 percent for 50 ppm, and 18.2 percent for 100 ppm. These lower amounts of fat for treated plants correspond with the decline in caloric content with increased 2,4-D treatment.

Sunflower Production

Plant growth following spray treatment was significantly affected. Mean change in height between 31 May and 15 June was 14.27 cm for controls and 17.22 and 8.20 cm for plants sprayed with 50 and 100 ppm of 2,4-D, respectively. Growth following the 100 ppm treatment was significantly ($P < 0.01$) less than growth of controls. The 50 ppm treatment seemed to increase growth, although this difference was not significant ($P > 0.10$).

By the end of the growing season there were no significant differences ($P_L = 0.4732$) in height between treated and untreated plants. Average heights were 99.73 cm for control plants and 99.77 and 106.27 cm for plants treated with 50 and 100 ppm of 2,4-D, respectively (Table 9, 21).

Table 7. Caloric content of sunflower and ragweed seeds collected from 2,4-D treated plants and from control plants in 1976. Caloric content was determined by bomb calorimetry.

<u>Species and Treatment</u>	<u>Mean cal/g $\pm$ Standard Error¹</u>
<u>Sunflower</u>	
Control	5852.00 $\pm$ 19.75 a
50 PPM	5933.50 $\pm$ 28.12 a
100 PPM	5924.06 $\pm$ 46.73 a
<u>Ragweed</u>	
Control	5651.20 $\pm$ 7.40 b
50 PPM	5524.84 $\pm$ 12.53 c
100 PPM	5618.20 $\pm$ 8.79 d

¹Means sharing a common letter are not significantly (P > 0.05) different.

Table 8. Composition of ragweed and sunflower seeds collected from 2,4-D treated plants during 1976.
Composition detected by proximate analysis.

Species	Treatment	Percent Composition					Digestible Carbohydrates
		Moisture	Protein	Ash	Fat	Fiber	
Ragweed	Control	4.7	22.1	3.1	17.6	32.4	20.1
Ragweed	50 PPM	4.8	25.8	3.3	16.0	31.8	18.3
Ragweed	100 PPM	4.8	25.1	3.2	16.4	32.3	18.2
Sunflower	Control	5.0	18.3	3.2	27.7	27.0	18.8
Sunflower	50 PPM	4.9	17.0	3.2	27.8	27.7	19.4
Sunflower	100 PPM	4.7	17.4	3.1	27.6	27.5	19.7

Table 9. Sunflower plant production in 1976 for controls and two 2,4-D treatment levels. Treatment sums of squares were partitioned into linear and nonlinear, and probabilities of a greater F value are given for each.

	Treatment Means			Probabilities	
	Control	50 PPM	100 PPM	P _{linear}	P _{nonlinear}
Final plant height (cm)	99.726	99.774	106.265	0.4732	0.6934
Total seeds/plant	154.374	195.224	308.912	0.0266	0.5687
Unpredated seeds/plant	142.036	181.001	285.802	0.0256	0.5792
Seed weight/plant	0.710	0.937	1.579	0.0215	0.5457
Total capitula/plant	2.797	3.086	4.395	0.0298	0.4308
Predated capitula/plant	0.484	0.485	0.596	0.3391	0.5986
Capitula predation (%)	14.755	13.585	11.943	--	--
Seed predation (%)	7.992	7.285	7.481	0.9921	0.8660

Total number of seeds produced per flower head were not significantly different ($P_L = 0.2632$) between treatments, although plants sprayed with 2,4-D produced slightly more seeds per flower head (52.96, 61.52, and 63.39 for controls, 50 and 100 ppm, respectively) (Table 10, 22). This slight increase in seeds per capitulum was probably due to a slight, insignificant ($P_L = 0.4813$ for diameter-1 and $P_L = 0.3564$ for diameter-2) increase in diameter of seed heads in treated plants. Two diameters were taken for each seed head due to their irregular shape. Control seed head diameters averaged 1.09 and 1.00 cm, plants sprayed with 50 ppm of 2,4-D had average seed head diameters of 1.13 and 1.04, and seed heads from plants sprayed with 100 ppm had average diameters of 1.14 and 1.05 cm. Diameters were significantly ($P < 0.01$) correlated with total seed production ($r = 0.8292$ and $r = 0.8104$ for diameter-1 and diameter-2, respectively) (Table 11).

Seed weight increased slightly with treatments (5.00, 5.18, and 5.53 mg/seed for controls, 50, and 100 ppm of 2,4-D, Table 10). Weight of seed per flower head was not significantly different ($P_L = 0.2237$), although weights increased slightly with application of 2,4-D (0.235 g/capitulum for controls and 0.287 and 0.301 g/capitulum for 50 and 100 ppm).

Average time of seed-ripening was delayed by 2,4-D treatment ($P_L = 0.0182$), suggesting a delay in flowering time (Table 10). Seed heads were categorized by week of seed-ripening, beginning with the week of 5 August as week 0. Mean ripening times for seeds from control plants was week 7.39 and from plants treated with 50 and 100 ppm of 2,4-D were weeks 7.58 and 8.19, respectively. (Week 7 is the week of 23 September, and week 8 is the week of 30 September.)

The number of flower heads produced per plant increased significantly ($P_L = 0.0298$) with treatment. Control plants produced an average of 2.80

Table 10. Sunflower production on a flower head basis in 1976 for controls and 2,4-D treatments. Treatment sums of squares were partitioned into linear and nonlinear, and probabilities of a greater F value are given for each.

	Treatment Means			Probabilities	
	Control	50 PPM	100 PPM	P _{linear}	P _{nonlinear}
Seed-ripe time	7.395	7.583	8.194	0.0182	0.4457
Diameter-1 (cm)	1.095	1.131	1.144	0.4813	0.8449
Diameter-2 (cm)	0.995	1.043	1.052	0.3564	0.7207
Total seeds	52.995	61.523	63.393	0.2632	0.6782
Unpredated seeds	49.001	56.927	58.875	0.2567	0.6908
Seed weight/head (g)	0.235	0.287	0.301	0.2237	0.6830
Weight/seed (mg)	5.00	5.18	5.53	--	--

flowers while plants treated with 50 and 100 ppm produced 3.09 and 4.39 flowers, respectively (Table 9).

With the increased number of flower heads per plant and the small, insignificant increase in number of seeds per flower head with increased level of 2,4-D, total seed production per plant increased linearly ($P_L = 0.0266$) with the 2,4-D dosage. Mean seed production for controls and the 50 and 100 ppm treatments were 154.37, 195.22, and 308.91 seeds per plant, respectively (Table 9). Number of unpredated seeds per plant also increased significantly ($P_L = 0.0256$) as the 2,4-D dosage increased. Mean numbers of unpredated seeds were 142.04 for control plants and 181.00 and 285.80 seeds for plants treated with 50 and 100 ppm of 2,4-D, respectively. Seed weight per plant also increased ($P_L = 0.0215$) with treatment, as expected since the number of seeds significantly increased with 2,4-D treatment and treated seeds weighed slightly more than untreated seeds. Control plants produced an average of 0.710 g of seed, plants treated with 50 ppm of 2,4-D produced 0.937 g of seed, and plants treated with 100 ppm of 2,4-D produced an average of 1.579 g of seed per plant.

Seed predation was not significantly affected by treatment (Table 9). Overall predation on seeds in 1976 was 7.63 percent (7.99 percent for controls, and 7.29 and 7.48 percent for 50 and 100 ppm, respectively). Loss of the entire flower head due to girdling of the flower stem and its subsequent breakage before seed-set took place accounted for the loss of 13.50 percent of all flowers (14.76 percent for controls, and 13.59 and 11.94 percent for 50 and 100 ppm of 2,4-D, respectively). Numbers of flowers predated per plant were not significantly different between treatments (0.48, 0.49, and 0.60 flowers per plant for controls, 50, and 100 ppm, respectively, $P_L = 0.3391$). Combining seed predation and flower girdling for controls shows 21.57 percent overall predation, 19.89 percent for 50 ppm, and 18.53 percent for 100 ppm.

Correlation analysis on a flower head basis showed that large flower heads produced large seed ($r = 0.4158$ and 0.4110 for diameter-1 and diameter-2, respectively, $P < 0.01$, Table 11). Weight per seed also increased with number of seeds produced per flower head ($r = 0.2501$, $P < 0.01$). Number of seeds per capitulum increased with time of ripening of seeds ($r = 0.2928$, $P < 0.01$). Capitula produced late in the year produce more seeds (Table 12). The later seed ripening time of treated sunflower plants (Table 10) probably contributed to the overall increase in seed production on a plant basis.

When compared on a plant basis, seed predation, calculated as predated seeds per total seeds, was negatively correlated with average time of producing seed ($r = -0.3881$, $P < 0.01$), average diameter of flower heads ($r = -0.1522$, $P < 0.01$), and total number of seeds produced ($r = -0.1151$, $P < 0.05$) (Table 13). This would indicate that seed predators get a higher proportion of the seeds when there are fewer seeds.

Predation of flowers followed the same pattern as seed predation. Flower predation was negatively correlated with average time of producing seed ($r = -0.3137$, $P < 0.01$), average diameter of capitula ($r = -0.3355$, $P < 0.01$), and number of flowers per plant ($r = 0.1300$, $P < 0.01$). This would indicate that the beetles responsible for this predation get a higher proportion of flower heads on plants with few, small flower heads.

Working with sunflowers, Gaines et al. (1974) defined reproductive effort as dry weight of reproductive tissue (flowers, heads, and seeds) per total dry weight of the plant. These data were not collected in this study, however another somewhat less accurate measure of reproductive effort would be weight of seeds per height of plant. Density of plants were fairly constant between treatments, therefore height of plant and biomass of a plant should be closely correlated, although microenvironmental factors would

Table 12. Mean seed production of sunflower capitula in 1976 by ripening time and treatment level.

Time	N	Total Seeds		Unpredated Seeds	
		(mean $\pm$ std. error)		(mean $\pm$ std. error)	
5 August					
Control	16	6.875	$\pm$ 3.513	2.438	$\pm$ 1.768
50 ppm	21	1.095	$\pm$ 0.740	0.333	$\pm$ 0.242
100 ppm	14	2.357	$\pm$ 1.492	1.143	$\pm$ 0.776
12 August					
Control	22	10.864	$\pm$ 2.810	5.364	$\pm$ 1.668
50 ppm	15	16.867	$\pm$ 3.008	10.533	$\pm$ 2.457
100 ppm	4	9.250	$\pm$ 9.250	7.500	$\pm$ 7.500
20 August					
Control	3	39.667	$\pm$ 37.684	38.333	$\pm$ 38.333
50 ppm	3	0.667	$\pm$ 0.667	0.667	$\pm$ 0.667
100 ppm	3	31.667	$\pm$ 18.187	29.333	$\pm$ 16.476
27 August					
Control	2	8.500	$\pm$ 8.500	8.000	$\pm$ 8.000
50 ppm	8	16.125	$\pm$ 7.952	11.250	$\pm$ 6.377
100 ppm	2	13.000	$\pm$ 13.000	13.000	$\pm$ 13.000
3 September					
Control	5	17.000	$\pm$ 17.000	14.600	$\pm$ 14.600
50 ppm	9	7.444	$\pm$ 2.759	6.444	$\pm$ 2.375
100 ppm	7	2.143	$\pm$ 1.122	0.714	$\pm$ 0.565
10 September					
Control	21	8.952	$\pm$ 3.585	8.143	$\pm$ 3.410
50 ppm	14	15.357	$\pm$ 9.176	11.429	$\pm$ 8.802
100 ppm	17	15.706	$\pm$ 5.508	10.882	$\pm$ 5.157
17 September					
Control	20	5.700	$\pm$ 2.696	5.050	$\pm$ 2.365
50 ppm	15	18.200	$\pm$ 7.819	16.800	$\pm$ 7.474
100 ppm	25	10.160	$\pm$ 3.675	9.080	$\pm$ 3.379
23 September					
Control	36	37.389	$\pm$ 8.496	35.250	$\pm$ 8.283
50 ppm	43	52.209	$\pm$ 7.555	46.581	$\pm$ 6.974
100 ppm	75	48.453	$\pm$ 7.584	45.187	$\pm$ 7.321
30 September					
Control	80	63.438	$\pm$ 7.669	58.588	$\pm$ 7.226
50 ppm	98	92.622	$\pm$ 8.867	87.082	$\pm$ 8.643
100 ppm	130	84.108	$\pm$ 6.171	77.477	$\pm$ 5.871
7 October					
Control	108	93.861	$\pm$ 6.733	86.824	$\pm$ 6.330
50 ppm	84	98.083	$\pm$ 7.361	91.143	$\pm$ 7.090
100 ppm	153	109.667	$\pm$ 5.709	103.320	$\pm$ 5.509
14 October					
Control	100	55.640	$\pm$ 6.141	51.730	$\pm$ 5.929
50 ppm	139	55.245	$\pm$ 4.760	51.367	$\pm$ 4.568
100 ppm	220	63.809	$\pm$ 4.232	58.402	$\pm$ 3.999

Table 13. Correlation coefficients of sunflower plant production parameters in 1976.

(1) Height	1.0000**									
(2) Ave. flowering time	0.3883**	1.0000**								
(3) Ave. diameter-1	0.5377**	0.5938**	1.0000**							
(4) Ave. diameter-2	0.5360**	0.5873**	0.9808**	1.0000**						
(5) Total seeds	0.6260**	0.2846**	0.4877**	0.4810**	1.0000**					
(6) "Good" seeds	0.6204**	0.2847**	0.4826**	0.4755**	0.9986**	1.0000**				
(7) Weight/plant	0.6111**	0.2603**	0.4651**	0.4616**	0.9778**	0.9740**	1.0000**			
(8) No. of flowers	0.5476**	0.2557**	0.3171**	0.3140**	0.7908**	0.7844**	0.7802**	1.0000**		
(9) % flower predation	0.0569	-0.3137**	-0.3355**	-0.3536**	-0.0497	-0.0473	-0.0395	-0.1300**	1.0000**	
(10) Seed predation	-0.0584	-0.3881**	-0.1522**	-0.1545**	-0.1151*	-0.1314*	-0.1025	0.0574	-----	1.0000**
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)

* - Significant at the 0.05 level.

** - Significant at the 0.01 level.

tend to influence it. Reproductive effort compared on this basis show significant differences among treatments ($P < 0.05$) for sunflower. Means were 5.93 mg/cm for controls and 7.51 and 11.11 mg of seed per cm of height for the 50 and 100 ppm treatments. This would indicate increased reproductive effort with 2,4-D treatment in sunflower.

Seed composition:

Bomb calorimetry showed no significant differences ($P = 0.2056$) in energy content of seeds between treatments. Control plants produced seeds containing 5852.00 cal/g while plants treated with 50 and 100 ppm of 2,4-D contained 5933.50 and 5924.06 cal/g, respectively (Table 7).

Proximate analysis showed that protein content was lower in treated seeds than in controls (18.3 percent for controls, and 17.0 and 17.4 percent for 50 and 100 ppm, respectively, Table 8). Moisture, fat, ash, and fiber content appeared very consistent among treatments, while digestible carbohydrates appeared to increase slightly with treatment (18.8 percent for controls, 19.4 and 19.7 percent for 50 and 100 ppm, respectively).

Seed color:

Comparison of seed coat color between treated and untreated plants showed no significant results, however color appeared to get darker with increased 2,4-D treatment ($P = 0.2185$) and time ($P = 0.1018$). Higher values for color indicate darker color on a yellow-gray scale: a value of 1 corresponds to a light tannish-yellow while a value of 9 is dark gray (almost black). Mean color of control seeds produced between 5 August and 10 September was 6.34 while for the same period, plants treated with 50 and 100 ppm of 2,4-D averaged 6.75 and 7.30, respectively (Table 14). Seeds produced later in the year were also darker. Mean color of seeds from control plants was 6.34 for the 5 August to 10 September period, 6.89 for the 17 September to

Table 14. Color of seeds from sunflower plants under three 2,4-D treatments and from differing seed ripening times. Ascending color values indicate darker seed coat color on a yellow-gray scale: 1 corresponds to light tannish-yellow, and 9 is dark gray.

<u>Time</u>	<u>Mean Seed Color</u>		
	<u>Control</u>	<u>50 PPM</u>	<u>100 PPM</u>
5 Aug - 10 Sept	6.34	6.75	7.30
17 Sept - 24 Sept	6.89	6.88	7.50
30 September	7.57	7.17	7.28
7 October	7.26	7.36	7.30
14 October	7.35	7.44	7.51

24 September period, 7.57 for 30 September, 7.26 for 7 October, and 7.35 for the 14 October period. Seeds from plants treated with 50 ppm of 2,4-D also appeared darker later in the year, while plants treated with 100 ppm were dark throughout the year (Table 14).

DISCUSSION

Spray Conditions

In attempting to compare the effects of one spray treatment to other spray treatments, many factors must be taken into consideration. Some of these are temperature, light, humidity, water condition of the plant (soil moisture), and spray-particle size. Temperature (18.3-29.4°C), humidity (100 percent), and soil moisture (32.77 percent for 30-40 cm depths) conditions in this study were near optimal for the maximum effect of 2,4-D (e.g., Muzik 1965 for temperature; Pallas 1958 for humidity; Basler et al. 1961 for soil moisture). Skies were clear for 48 h following spray treatment, which many authors have shown to decrease plant reaction to 2,4-D spray compared to cloudy conditions (e.g., Mitchell and Brown 1946). Particle size was not measured in this study, but the spray particles tended to bead up on the plant. I believe particle size was fairly large, which would tend to reduce the effect of 2,4-D (Ennis and Williamson 1963, Way 1969, McKinlay et al. 1972).

The two reasons given by McKinlay et al. (1972) for smaller particles having a greater effect (larger droplets killing leaf cells directly under them and leaf area contacted by a given volume increasing with smaller droplets) may, however, not have as great an effect here because of the dew present. A spray particle landing on a dew drop would tend to increase the surface area contacted by 2,4-D, as well as dilute the 2,4-D solution to a point where it may not kill the leaf cells directly under it. All factors taken together, large spray particle size here probably did not reduce the effect of the 2,4-D significantly.

Seed Production

Results of seed production in 1975 and for controls in 1976 were considerably below those found for sunflower and ragweed by Stevens (1932)

in North Dakota. Using one "average" plant relatively free from competition, he found weight per sunflower seed of 6.58 mg, and 72,000 seeds per plant. Seed weights in the present study were 5.47 and 5.00 mg for 1975 and 1976, respectively, and production was 105.61 seeds per plant in 1975 and 154.37 seeds per plant in 1976. For ragweed, Stevens found 3380 seeds per plant and 3.95 mg per seed in one "average" competition-free plant. In my study, production was 36.58 and 94.39 seeds per plant in 1975 and 1976, respectively. Weight per seed was 3.15 mg in 1975 and 2.27 mg in 1976. Bishop and Spinner (1946) found an average weight of ragweed seeds of 2.81 g per plant in Connecticut cornfields, also considerably higher than results from my study (0.113 g in 1975 and 0.234 g in 1976).

Dry conditions may be one reason for the relatively low seed production in the ragweed area. The soil type (Benfield-Florence complex) is relatively dry (U.S. Dept. Agric. Soil Cons. Service 1975), and its south-facing slope make it even drier. Both 1975 and 1976 were dry years, although August and September 1975 were fairly wet. Moisture throughout the growing season is important in seed production, although moisture at the time of seed-filling is probably most important. Rainfall between 1 August and 1 October 1975 was 131 percent of the 1931-60 means (Bark 1963). In 1976, rainfall between 6 May and 6 October was only 49 percent of normal (using data from the ragweed study area). (See rainfall and soil moisture data, Appendix.) In addition, plowing of this area for a fire break had caused erosion of most of the topsoil and the soil present was probably very low in nutrients.

In the sunflower area, low production may also be due to the dry conditions and competition. Succession is now favoring grasses and sunflower, an early successional plant, is being slowly out-competed. Early successional plants generally have a low root/shoot ratio, making them poor competitors

for water and nutrients (Ricklefs 1973). More energy can therefore be allocated to reproduction. When competition forces early successional plants to generate a higher root/shoot ratio to compete for water and nutrients, reproductive effort must decline. This could have reduced seed production of sunflower plants in the study area.

The effects of 2,4-D on seed production in sunflower and ragweed plants differed. Treated sunflower plants produced significantly more seeds than controls, while treated ragweed plants produced slightly fewer seeds. Several studies have been done on 2,4-D treated plants prior to flowering, with variable results. Weaver (1946) found that spraying young (6-8 inches high) soybeans with low levels of 2,4-D did not reduce yield. One fourth pound per acre of 2,4-D did not reduce ragweed seed production in Michigan when plants were sprayed at an early stage of growth (Jameson and Wheeler 1951). Greenshields and White (1954) and Greenshields and Putt (1958) decreased seed production in cultivated sweet clover and cultivated sunflower, respectively, by exposing young plants to drift of 2,4-D.

Several studies have shown an increase in seed production with low levels of 2,4-D in early stages of plant growth. Wedding et al. (1956) showed increased yields of lima and pole beans by the application of 10 to 20 ppm of 2,4-D when the plants were two weeks old. Miller et al. (1962) found increased production of field beans (20 percent) with low levels (up to 10 ppm) of 2,4-D. When low levels of 2,4-D were put in irrigation water, cotton seed production increased slightly (Arle 1954). Treatment of the soil with 2,4-D in the fall increased seed production slightly in cultivated sunflowers (Vasil'yev and Annenkova 1964). This would seem to simulate low levels of 2,4-D. They found similar results in coriander, castor beans, and soybeans, but all these results were due at least in part to decreased weediness of plots.

Based on previous studies, it appears that very low levels of 2,4-D may increase growth of a plant and increase seed production. The actual level of 2,4-D to produce this effect varies with the species involved, climatic factors, and the physiological condition of the plant. Higher levels, of course, decrease production, while still higher levels kill the plant. This relationship is shown in Figure 1. For sunflower under the conditions it was sprayed in this study, 100 ppm is still increasing production. For ragweed under the conditions in this study, 50 ppm decreased production. Lower levels may increase production in ragweed, but these levels were not tested in this study. Climatic and physiological factors would shift the curve to the right or left, or up or down. For example, low humidity would lessen the effect of 2,4-D and shift the curve to the right.

Average seed-ripening time was later for treated sunflowers in this study. Plants treated with 100 ppm of 2,4-D produced ripe seeds an average of almost one week later than controls. Plants sprayed with 50 ppm were intermediate. As time of seed-ripening became later in this study, seed production per capitulum increased for control and treated plants. Many authors have found a similar delay in flowering time with low levels of 2,4-D using different plants. Miller et al. (1962) found delayed maturation of field bean seeds when sprayed with 5 ppm of 2,4-D or more. Delayed maturation was also found in cotton (Arle 1954), turnip rape (Betts and Ashford 1976), and sweet clover (Greenshields and Putt 1954). Delayed maturation may be the reason for the reduced seed production of ragweed with 2,4-D treatment. Due to unknown reasons (possibly the dry conditions) ragweed bloomed only 7 to 10 days before the first killing frost. Had there been sufficient moisture or time before the first killing frost, production for treated plants may have equalled or surpassed controls.

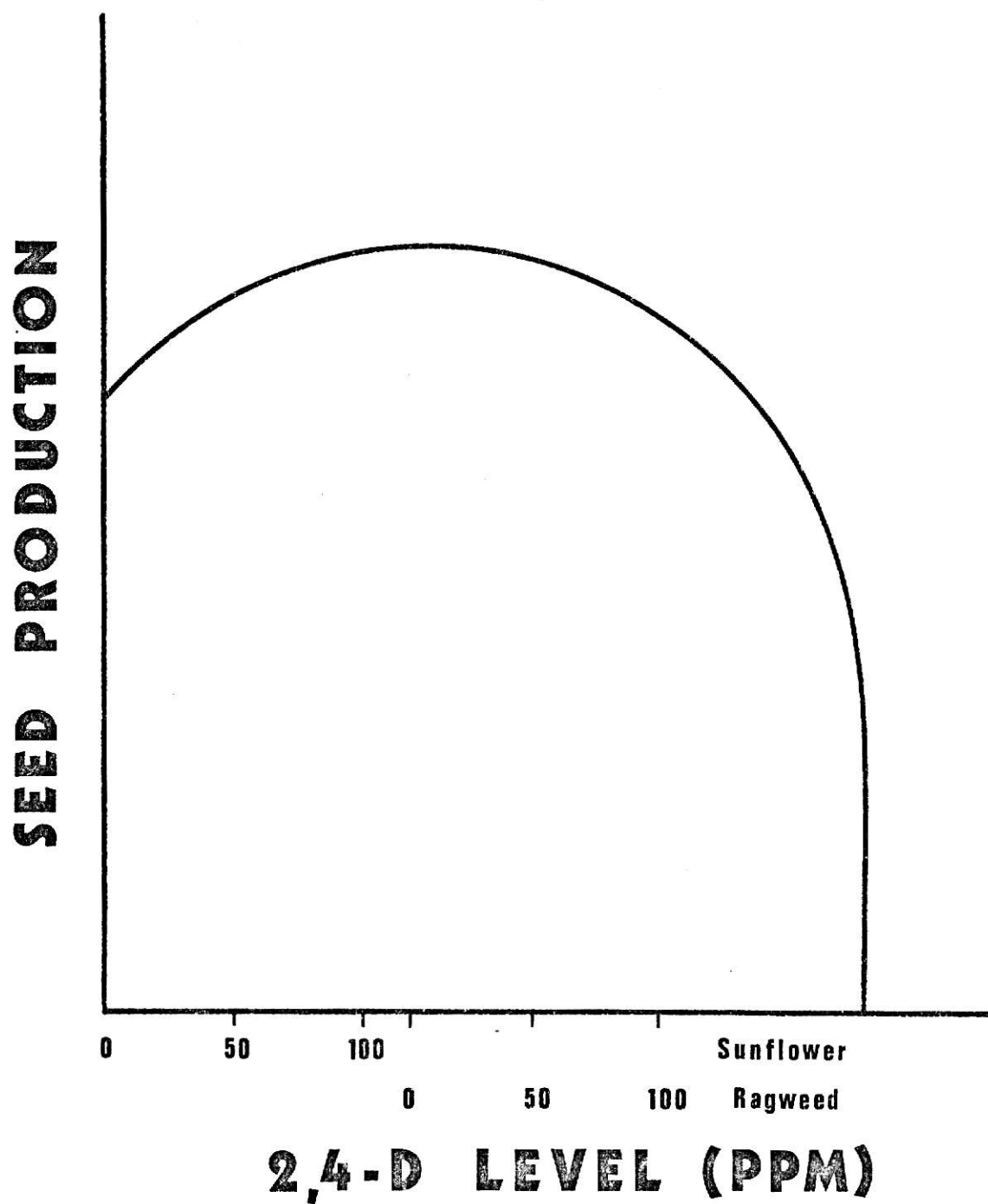


Fig. 1. Hypothetical curve estimating the effect of 2,4-D on seed production.

There was no significant difference in weight per seed with increased treatment level, however, there was a trend towards an increased seed weight with treatment. For ragweed, weights per seed averaged 2.27, 2.33, and 2.39 mg for controls, 50, and 100 ppm, while for the same treatments in sunflower mean weights were 5.00, 5.18, and 5.53 mg per seed. Increased weight with 2,4-D treatment has been found in soybeans by Basnet et al. (1972). Betts and Ashford (1976) found increased size of turnip rape seeds when sprayed at the time of flower formation or later. Taylor (1947) found increased weights of bean plants and corn tops when these crops were planted 3-5 weeks after the soil was treated with 2 lbs/acre (2.24 kg/ha) of 2,4-D. This would seem to simulate low levels of 2,4-D. Conversely, seed weight was reduced due to early treatment of cultivated sunflowers with 2,4-D (Greenshields and Putt 1958).

While there was a slight increase in seed number per flower head with treatment, the main reason for an increase in production for sunflower was a greater number of flowers on treated plants. Cultivated sunflower produce only one seed head, and this could at least partially explain the reason Greenshields and Putt (1958) found reduced seed production on treated sunflower plants. They, however, sprayed under different climatic conditions and do not state the levels of 2,4-D used.

Results of the proximate analysis for sunflower and ragweed controls were similar to those of Korschgen (1964) in Missouri except for fat content of ragweed. Fat content of ragweed seeds in my study were higher (21.0 and 17.6 percent for 1975 and 1976 controls, respectively) than in Missouri (12.52 percent).

Studying wheat, Klingman (1950) found no difference in protein content when 2,4-D was applied at a rate that did not affect seed production. When

higher levels were used and seed production was reduced, protein content of the wheat increased significantly. In the present study, protein content of ragweed increased with a decrease in production due to 2,4-D (22.1 percent for controls and 25.8 and 25.1 percent for 50 and 100 ppm of 2,4-D, respectively). Sunflower, however, showed a slight decrease in protein content with increased 2,4-D treatment (18.3 percent for controls, 17.0 percent for seeds from plants treated with 50 ppm, and 17.4 percent for the 100 ppm treatment). Sunflower seed production increased, and this could explain the decrease in protein levels. It appears plants can take one of two strategies: produce few seeds and invest a lot of energy in each, or produce many seeds and invest less energy per seed. These results are in agreement with those found by Klingman.

No reports in the literature were found concerning the effect of 2,4-D on caloric content of seeds. In a related study, however, Betts and Ashford (1976) found lower quantities of oil from turnip rape treated with low levels of 2,4-D. This would tend to reduce caloric content. Lower caloric content of seeds from treated ragweed plants is probably due to a decrease in fat content with 2,4-D treatment. Sunflower showed no significant differences in caloric content between treatments, and there was virtually no difference in fat content, as well.

Caloric content for untreated sunflowers (5915.48 and 5852.00 cal/g for 1975 and 1976) are in fairly close agreement with other results found in Kansas (Robel and Harper 1965, Robel 1972). Johnson and Robel (1968) found lower values (5573 cal/g) for sunflower. Kendeigh and West (1965) found ragweed values of 5286 cal/g, considerably less than those found for controls in this study (5843.96 and 5657.20 for 1975 and 1976, respectively). These differences might be explained by microenvironmental factors, especially in

seeds with high fat content such as sunflower and ragweed (Johnson and Robel 1968).

Seed Color

The trend toward darker sunflower seeds with 2,4-D treatment could have an effect on seed eaters. Several authors (Kalmbach 1943, Kalmbach et al. 1946, Allen and Clark 1968, Pank 1976) have shown that color of seeds affects avian consumption. Pank (1976) found lowered consumption by California quail (Lophortyx californicus) fed charcoal-dusted darker seeds. Texture of the seeds also changed, however, and this may have had an effect. The color change in this study was slight, but could have an effect on bobwhite consumption.

Seeds from control sunflower plants were lighter in color early in the year than late in the year. Seed color probably has adaptive value. In Kansas, soil moisture is generally higher in late September and October than in August and early September, and moist soils are usually darker than dry soils. By producing lighter seeds when soil is dry and dark seeds when soil is moist, predation of seeds by predators which key on sight may be reduced. By producing some light and some dark seeds throughout the year, problems due to inter-seasonal variation in moisture can be minimized. Plants treated with low levels of 2,4-D produce dark seeds throughout the growing season, and there was less variation in color of seeds from 2,4-D treated plants than from controls. This may increase the probability of a seed being consumed by a granivore and reduce the number of seeds available for germination the following spring.

No other reports could be found showing 2,4-D-induced darker seeds. While two-dimensional paper chromatography showed no difference in phenolic compounds in sunflower seeds, quantities of phenolics may have been different

between 2,4-D treatment levels. Proximate analysis and calorimetric differences were discussed earlier but may not be the entire story. It is possible that this change of color may affect seed germination or be associated with another physiological process. More work should be done to determine whether this change in color induced by 2,4-D will be associated with abnormal plant growth or reduce bobwhite consumption.

CONCLUSIONS

Under the conditions of this study, the following conclusions seem justified:

1. Levels of 50 and 100 ppm of 2,4-D sprayed at 0.1 liters/m² (or 1/5 to 1/10 of the recommended application rates) did not significantly affect ragweed seed production or weight of seed. Plant height was significantly reduced with treatment. Caloric content of seeds from treated plants was significantly lower than controls. Proximate analysis of ragweed seeds showed higher protein and lower fat content in treated seeds.
2. Levels of 50 and 100 ppm of 2,4-D sprayed at 0.1 liters/m² significantly increased the number of seeds produced by sunflower plants. Treated plants produced more seed and a heavier total weight of seed per plant than controls. Flowering time was delayed with treatment, and capitula produced late in the year produced more seed than capitula produced early in the year. Significant differences in caloric content were not found. Proximate analysis of sunflower seeds showed a slight reduction in protein content of seeds from treated plants.
3. Although not significant, seeds from sunflower plants treated with 2,4-D appeared to be darker in color than controls, and seeds produced late in the year appeared darker than seeds produced early in the year.
4. Agricultural drift of 2,4-D at the levels used in this study may be beneficial to bobwhite. Sunflower seed production was doubled and time of flowering was delayed. This may mean more seeds available to bobwhite later in the year when seeds are most needed. Germination of treated seeds should be tested.

SUMMARY

In order to determine the effects of 2,4-D drift on bobwhite food sources, 30 2x3-m plots of sunflower and 30 2x3-m plots of ragweed plants were demarcated. Adjacent plots were grouped into blocks of three plots each, and within each block the three treatments (control, 0.050 kg acid equivalent of 2,4-D/ha, and 0.100 kg acid equivalent of 2,4-D/ha) were randomly assigned. The two 2,4-D levels were sprayed at 50 and 100 ppm at a rate of 1/10 liters/m². Seed production was determined by collecting sunflower seed heads and entire ragweed plants shortly after the seeds ripened.

Seed production of ragweed plants treated with 2,4-D did not differ significantly from controls, although it was reduced slightly. Treated plants were significantly shorter in height than control plants, and this may have accounted for the slight reduction in seed production in treated plants. Proximate analysis showed that protein content of seeds from treated plants was higher than for controls, but there was very little difference in protein content between 2,4-D treatments. Fat and digestible carbohydrate levels were lowered in seeds from 2,4-D-treated plants than in controls. This reduction in fat and digestible carbohydrate levels may account for the significant reduction in caloric content of seeds from treated plants compared to controls.

Sunflower seed production in treated plants significantly increased with 2,4-D treatment. While the number of seeds produced per capitulum increased insignificantly with treatment, the number of flower heads produced by plants treated with 100 ppm of 2,4-D was almost twice as great as the number produced by controls. Treated plants produced more seed by producing more flowers. The plants treated with 100 ppm of 2,4-D produced approximately

twice as many seeds as controls, while plants treated with 50 ppm were intermediate. Proximate analysis of sunflower seeds showed that protein content of seeds from treated plants was slightly lower than in controls. Digestible carbohydrate levels were slightly higher in seeds from treated plants. There were no significant differences in caloric content among the treatments.

Color of sunflower seed coats was observed to be darker in treated than in untreated seeds. Seed color was quantified by comparing seeds to a yellow-gray color chart which was numbered from light yellow to almost black. Because this difference was not noted until after seeds from capitula and plants were mixed together, only means of seed color could be compared. Color differences tested in this manner were not significant. Seeds produced late in the year appeared darker than seeds produced early in the year, but this also was not significant.

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APPENDIX

Table 15. Rainfall data for the sunflower and ragweed study areas in 1975 beginning 1 August 1975. Values given are in centimeters.

Date	Sunflower	Normal for month (Bark 1963)	Ragweed
13 August	3.68		2.29
14 August	2.16		2.03
17 August	1.65		2.79
25 August	0.38		0.76
26 August	Trace		Trace
28 August	2.41		1.14
29 August	0.76		0.51
Total for August	11.04	10.36	9.52
5 September	6.22		5.33
11 September	5.84		2.79
14 September	0.13		0.13
18 September	Trace		Trace
19 September	Trace		Trace
28 September	0.51		1.14
29 September	0.25		0.13
Total for September	12.95	8.00	9.52
1 October	1.78		2.16

Table 16. Biweekly soil moisture data from three depths in the sunflower study area in 1975. Values are in percent.

Date	Depths		
	10-20 cm	30-40 cm	50-60 cm
31 July	9.49	16.77	16.17
14 August	26.15	20.45	20.80
28 August	20.89	19.65	19.36
13 September	31.24	31.91	26.96
25 September	25.89	31.75	26.72
9 October	26.96	30.84	26.50

Table 17. Weather data for the week of spray application of 2,4-D. Data were collected by the KSU Department of Physics.

<u>Date</u>	<u>High Temp.</u>	<u>Low Temp.</u>	<u>Precipitation</u>	<u>Humidity (6 am)</u>
31 May	80°F (27°C)	53°F (12°C)	0	100%
1 June	85°F (29°C)	55°F (13°C)	0	100%
2 June	83°F (28°C)	60°F (16°C)	0	100%
3 June	82°F (28°C)	59°F (15°C)	0	86%
4 June	82°F (28°C)	52°F (11°C)	0	68%
5 June	82°F (28°C)	60°F (16°C)	0	98%
6 June	83°F (28°C)	61°F (16°C)	Trace	98%

Table 18. Weekly soil moisture data from three depths in the sunflower study area in 1976. Values are in percent.

Date	Depths		
	10-20 cm	30-40 cm	50-60 cm
13 May	27.82	31.92	31.23
20 May	27.90	31.21	28.43
27 May	32.56	33.65	30.68
1 June	27.11	32.77	28.75
3 June	22.56	32.49	27.86
10 June	16.56	29.72	28.68
17 June	14.23	27.10	26.99
24 June	26.74	28.50	25.70
1 July	24.81	27.42	25.26
8 July	22.09	28.42	26.85
15 July	14.78	22.36	21.43
22 July	15.43	23.75	22.74
29 July	15.13	21.23	20.60
5 August	17.72	19.68	19.04
12 August	11.96	18.68	17.94
19 August	11.31	18.19	17.13
26 August	4.18	14.56	16.84
2 September	Not taken	Not taken	Not taken
9 September	10.61	15.29	17.19
16 September	13.63	18.38	16.74
23 September	9.73	18.19	16.51
1 October	10.73	16.34	16.13
7 October	19.50	17.60	17.91
14 October	15.04	19.45	18.14

Table 19. Rainfall data for the sunflower and ragweed study areas in 1976 beginning 6 May 1976. Values given are in centimeters.

Date	Sunflower	Normal for month (Bark 1963)	Ragweed
11 May	0.51		0.51
15 May	3.05		2.84
22 May	0.76		0.76
23 May	2.29		3.94
26 May	0.56	9.12	0.51
Total for May	7.17		
10 June	Trace		Trace
11 June	Trace		Trace
14 June	Trace		Trace
17 June	2.24		3.12
23 June	4.27		4.27
27 June	0.20		0.33
29 June	3.40	13.28	3.30
Total for June	10.11		
15 July	0.13		0.13
17 July	1.91		1.91
27 July	0.86	9.70	0.30
Total for July	2.90		
5 August	0.18		0.18
14 August	0.13	10.36	0.13
Total for August	0.31		
8 September	0.38		0.38
13 September	0.30		0.51
15 September	1.78		2.03
22 September	0.25		0.51
25 September	1.66	8.00	1.45
Total for September	4.37		
4 October	0.64		0.64
5 October	0.76		0.76
6 October	Trace		Trace

Table 20. Source of variation, degrees of freedom and mean squares of two-way analysis of variance for ragweed plants; 1976.

Source	df	Mean Squares				Seed weight/ plant
		Height	Branches	Total Seeds	"Good" seeds	
Block	9	2467.710	9.876	50885.85	47626.03	0.4132
Treatment (linear)	1	1245.302	0.436	20610.09	19162.09	0.0758
(nonlinear)	1	13361.474	12.784	59742.54	56871.25	0.3368
Error (Interaction)	18	414.931	14.569	55043.32	53094.22	0.4102
Residual	717	138.712	4.595	11095.32	10669.06	0.0814

Source	df	Mean Squares	
		Weight/seed	Seed Predation
Block	9	0.0429	0.0054
Treatment (linear)	1	0.0174	0.0001
(nonlinear)	1	0.0001	0.0031
Error (Interaction)	18	0.0316	0.0103
Residual	717	0.0055	0.0040

Table 21. Source of variation, degrees of freedom and mean squares of two-way analysis of variance for sunflower plants in 1976.

Source	df	Mean Time of		Mean Squares		
		Height	Seed Set	Diameter-1	Diameter-2	Total seeds
Block	9	14440.91	32.445	0.5751	0.4665	520749.56
Treatment (linear)	1	3121.84	0.070	0.0696	0.0931	1743387.46
(nonlinear)	1	933.07	34.945	0.0000	0.0000	100828.35
Error (Interaction)	18	5814.36	7.726	0.3166	0.2741	299126.25
Residual	408	833.66	6.837	0.1384	0.1189	128510.75

Source	df	Mean Squares				
		"Good" Seed	Seed Weight/ Plant	No. Flowers	No. Pred. Flowers	Seed Pred.
Block	9	426498.25	23.410	58.879	2.301	0.0183
Treatment (linear)	1	1508819.88	55.100	186.315	0.913	0.0000
(nonlinear)	1	81267.40	3.295	21.896	0.272	0.0007
Error (Interaction)	18	254720.38	8.688	33.712	0.947	0.0254
Residual	408	112949.38	3.829	11.777	1.001	0.0240

Table 22. Source of variation, degrees of freedom and mean squares of two-way analysis of variance for sunflower seed heads, in 1976.

Source	df	Mean Squares			
		Time	Diameter-1	Diameter-2	Total Seeds
Block	9	41.394	1.4914	1.3487	17594.68
Treatment (linear)	1	129.475	0.4832	0.6556	22095.65
(nonlinear)	1	11.659	0.0368	0.0965	2956.33
Error (Interaction)	18	19.180	0.9345	0.7319	16563.36
Residual	1482	5.712	0.1690	0.1472	4458.49
					4092.88

Source	df	Mean Squares	
		Seed weight/head	
Block	9	1.5004	
Treatment (linear)	1	0.8745	
(nonlinear)	1	0.0949	
Error (interaction)	18	0.5508	
Residual	1482	0.1471	

THE EFFECTS OF LOW LEVELS OF 2,4-D ON SUNFLOWER
AND RAGWEED SEED PRODUCTION IN NORTHEASTERN KANSAS

by

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The purpose of this study was to determine the effects of drift-like levels of 2,4-D on bobwhite (Colinus virginianus) food sources. Sunflower (Helianthus annuus) and ragweed (Ambrosia artemisiifolia) are two important seeds for bobwhite, and study areas with high densities of these plants were located within the Fort Riley Military Reservation, 11 km southwest of Manhattan, Kansas.

Thirty 2 x 3-m plots of each species were grouped into blocks of three plots each. Within each block, the three treatments (controls, 0.050 kg acid equivalent (A.E.)/ha, and 0.100 kg A.E./ha) were randomly assigned. The two 2,4-D levels were sprayed at 50 and 100 ppm at a rate of 0.1 liters/m² when sunflower and ragweed plants averaged 2-4 pairs of leaves. Seed production was determined by collecting sunflower seed heads and entire ragweed plants shortly after the seeds ripened.

Seed production of ragweed plants treated with 2,4-D was reduced slightly, but this difference was insignificant. Treated plants were significantly shorter in height than control plants, and this may have accounted for the slight reduction in seed production in treated plants. Proximate analysis showed that protein content of seeds from treated plants was higher than for controls, but there was very little difference in protein content between 2,4-D treatments. Fat and digestible carbohydrate levels were lower in seeds from 2,4-D treated plants than in controls. This reduction in fat and digestible carbohydrate levels may account for the significant reduction in caloric content of seeds from treated plants compared to controls.

Sunflower seed production in treated plants significantly increased with 2,4-D treatment. The number of seeds produced per capitulum increased insignificantly with treatment, however the number of flower heads produced

by plants treated with 100 ppm of 2,4-D was almost twice as great as the number produced by controls. Treated plants produced more seed by producing more flowers. The plants treated with 100 ppm of 2,4-D produced approximately twice as many seeds as controls, while plants treated with 50 ppm were intermediate in seed production. Proximate analysis of sunflower seeds showed that protein content of seeds from treated plants was slightly lower than in controls. Digestible carbohydrate levels were slightly higher in seeds from treated plants. There were no significant differences in caloric content among the treatments.

Color of sunflower seed coats was observed to be darker in seeds from treated plants than in seeds from untreated plants. This difference was not noted until after seeds from capitula and plants were mixed together, therefore only means of seed color could be compared. Between treatment color differences tested in this manner were not significant ($P = 0.2185$). Seeds produced late in the year appeared darker than seeds produced early in the year, but this also was not significant ($P = 0.1018$).