# THE USE OF FACTORIAL EXPERIMENTS TO DETERMINE THE SENSORY PERCEPTION OF VARIOUS BAIT FACTORS BY *Musca domestica* L.

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THE USE OF FACTORIAL EXPERIMENTS TO DETERMINE THE

SENSORY PERCEPTION OF VARIOUS BAIT

FACTORS BY *Musca domestica* L.

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## PREFACE

The author became aware of the need for better methods of determining the sensory perception of various bait factors by the house fly, *Musca domestica* L., while working with baits during the summer and fall of 1961. After conferring with Dr. D. E. Howell, Professor and Head of the Department of Entomology, it was decided that a study of this nature would make an interesting and worthwhile thesis problem.

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# INTRODUCTION

The house fly, Musca domestica L., ranks high in importance among arthropods of medical and veterinary importance. This is due to a variety of factors. The cosmopolitan distribution, type of mouthparts, filthy habits, and rapid rate of reproduction create high vector potentialities for a wide range of diseases. The annoyance to man and his domestic animals is another factor which should be considered in the evaluation of the importance of this pest. Certainly, the unsightly presence of large numbers of house flies and their specks is not a problem to be overlooked. All of these factors indicate that the house fly is worthy of considerable investigation.

One objective of the investigations reported in this thesis was to improve experimental methods of determining the sensory perception of various bait factors by the house fly. Uniformity trials and preliminary observations were made with regard to both laboratory and field experimentation. This preliminary type of work brought about the use of new equipment and methods as compared to previous experimentation by other workers. This also allowed a more intelligent choice of statistical methods to be employed in the various tests.

Another objective of the experimentation was to determine the effectiveness of several fly bait constituents when subjected to various experimental conditions. Also, by the use of appropriate statistical methods, the interactions among the effects of the various factors and their types and levels were considered. Factorial experiments were used in investigations of interactions. It was thought that information would be best obtained by testing all combinations.

## REVIEW OF LITERATURE

EARLY CONTROL WITH BAITS.--Some of the first chemical controls used against the house fly, *Musca domestica* L., were poisoned baits. Weiss (1912) indicated that King's yellow, Quassia, and corrosive sublimate were used around 1800 to prepare a bait for house flies in England. In the early 1900's, Smith (1911), Morrill (1914), and Buck (1915) reported good results with formalin baits containing sugar, milk, or bread as attractants. Morrill also reported that ethyl alcohol was both attractive and poisonous to house flies when mixed 1 part to 10 parts of water. Fenton and Bieberdorf (1936) found a formalin bait containing milk, water, and molasses as attractants to be helpful in reducing fly populations.

CHEMICAL ATTRACTION.--Attraction mechanisms have been an area of fruitful research for some time. Many factors seem to play a role in the attraction of house flies to chemicals. Chemicals which attract the house fly, and which might be used to advantage in control, have been the subject of several investigators. Richardson (1916a) and (1916b) concluded that house flies are attracted by ammonia from fermenting organic substances. He found that ammonia attracts a preponderance of females which could be induced to oviposit upon certain fermenting organic substances. Richardson conceded, however, that the attraction response is complicated by other factors.

The response of the house fly to certain foods and fermentation products was investigated by Richardson (1917). He concluded that various carbohydrates such as glucose, fructose, maltose, lactose, sucrose, starch, and dextrin were not very attractive to house flies. Sucrose was consistently a poor bait. His experiments indicated that aqueous solutions of wheat flour and molasses to which sodium arsenite and amylic [sic] alcohol were added have considerable value as poisoned baits for house flies.

Speyer (1920) found that essential oils are unattractive to the house fly. Some of them evoked negative chemotropic stimuli, these being oil of *Pinus sylvestris* L., orange oil, lemon oil, citronella oil, oil of juniper berries, and possibly camphor oil. Also, some such as cedar oil, eucalyptus oil, and oil of bitter almonds were inactive in raising stimuli.

Richardson and Richardson (1922) could not definitely establish whether or not carbon dioxide acted as an attractant for house flies. They concluded, however, that carbon dioxide did not induce oviposition.

Laake et al. (1931) found that several materials were attractive to the house fly. These were bromoform, ethyl mercaptan, chloroform, butyraldehyde, and formaldehyde.

Chemicals have also been tested by means of an olfactometer (Wieting and Hoskins, 1939). Their results showed that mixed groups of house flies having a sex ratio of approximately unity were attracted to ammonia at a concentration of 0.012%, but were strongly repelled at concentrations greater than 0.03%. Carbon dioxide had no appreciable effect up to 2%. Ethyl alcohol attracted feebly at 0.012% and repelled above 0.05%. They also reported that females were more strongly attracted than males to ammonia.

Two workers, Vanskaya (1941) and Yates (1951), found that the addition of ammonium carbonate to baits improved their attractiveness. This material also stimulated oviposition. Several investigators reported the testing of large numbers of chemicals as attractants for house flies. Dethier, Hackley, and Wagner-Jauregg (1952) indicated that a 0.013 molar solution of iso-valeraldehyde was superior in its attraction of Musca spp. to all other materials tested. Also, extensive tests were made by Brown, West, and Lockley (1961) in which a large number of materials were considered in an olfactometer; then some of the better ones were tested in a closed room and in the field. They concluded that no single compound was so effective that its attractiveness could not be enhanced by admixture. The most attractive material consisted of a combination, in aqueous solution, of 5% malt extract, 0.5% ethyl alcohol, 0.02% skatol, and 1% acetal.

FLY FACTOR.--Various researchers mentioned a "fly factor" in reports on attractant studies. Barnhard and Chadwick (1953) concluded from their data that flies which visited a bait contributed to it some substance which enhanced its attractiveness to the species. They found that an attractive material soluble in 95% ethyl alcohol, but much less in ether or acetone, could be extracted from the flies.

Dethier (1955) suggested that flies which escaped from traps left a volatile contaminant on the outside of the trap entrance, and that flies within traps generated something which reacted with sugar to produce an attractive volatile brew. Furthermore, they stated that the contaminant is not species-specific, and that flies produced on their legs appreciable amounts of invertase which hydrolyzed sucrose, melezitose, and raffinose. Later, Acree et al. (1959) reported that in their tests only 10 to 15% of the flies were attracted to fed-on sucrose. This indicated that the fly factor is a low-order attractant. Also, these workers indicated that no evidence was found for a volatile chemical reaction product of sucrose which had appreciable attractiveness.

TARSAL STIMULATION.--Two sugars, sucrose and levulose, were used as stimulators of tarsal segments of the house fly (Deonier and Richardson, 1935). They found the tarsal segments to be sensitive to solutions of sucrose and levulose. It was assumed that the response to the sugars resulted from stimulation of chemoreceptor organs.

COLOR ATTRACTION.--The color of surfaces to which house flies are attracted has been a subject of investigation for many years. An early worker, Lodge (1918), found that house flies did not show any color preference. Contrasting results were found by Freeborn and Berry (1935). They used a checkerboard on which 13 different colors were painted, each surrounded by a 3-inch border of white. The most attractive colors in their test were dark blue, dark red, light grey, and canary yellow. Other colors in their order of attractiveness were orange, aluminum, jade green, light blue, white, coral, foam green, ivory, and primrose.

Food packages wrapped in various colors were tested by Harsham (1946) in a Peet-Grady chamber. The number of fly specks on the wrapper indicated that aluminum was more attractive than other colors. Other colors in their order of attractiveness were light red, strawberry red, orange, dark yellow, lime green, light yellow, and purple.

Waterhouse (1948) has done the most extensive amount of work on the attractiveness of different colors to house flies. His report stated that it had been clearly established that house flies have a definite order of preference for surfaces of various colors. Furthermore, such a preference may be either for the color of the surface, or the intensity of light reflected, or a combination of both factors. This work also suggested a correlation between the amount of light reflected by a painted surface and its attractiveness to flies. The lighter colors were less attractive than the darker colors. Waterhouse concluded that dark colors might be used to attract house flies to treated surfaces.

Howell (1961) found that, of a variety of colored baits, tan baits killed the most flies. Yellow, brown, and pink were more attractive than uncolored baits. Black, dark blue, red, and deep orange were the least attractive colors for house fly baits.

CONTROL WITH BAITS.--Resistance to DDT (dichloro diphenyl trichloroethane) and other chlorinated hydrocarbon insecticides has shifted emphasis to control with organic phosphates. Simultaneously, renewed interest has developed in regard to the use of baits for house fly control. Bruce (1953) proposed a new technique in the control of the house fly. He suggested a bait prepared from karo syrup and Bayer L 13/59 (dimethyl (2,2,2-trichloro-1-hydroxyethyl) phosphonate). This mixture was painted on various surfaces in fly infested areas and allowed to harden. Good control was reported over long periods of time. Bruce conceded that the bait might be improved with the addition of an attractant. The use of liquid baits is also mentioned in the same year by other authors (King, Guyer, and Ralston, 1953; Price, 1953; Gahan, et al., 1953; and Thompson, et al., 1953).

Gahan, Wilson, and McDuffie (1954a) reported that Diazinon (O,O-diethyl O-(2-isopropyl-6-methyl-4-pyrimidyl) thiophosphate) and Bayer L 13/59 were more toxic than malathion (O,O-dimethyl dithiophosphate of diethyl mercaptosuccinate) in an aqueous solution containing 10% molasses. Bayer L 13/59 reduced the attractiveness of baits slightly, and certain concentrations of malathion markedly reduced their acceptability.

Dry sugar baits containing 0.1% malathion, Diazinon, or Bayer L 13/59 gave 99% kills of flies in 16 hours, and higher concentrations gave faster kills. Baits stored for 1 month showed no loss of toxicity (Gahan, Wilson, and McDuffie, 1954b). Langford, Johnson and Harding (1954), and Johnson (1954) indicated good results with malathion, Diazinon, and Bayer L 13/59 baits. Johnson reported that initial kills were not as rapid with dry baits as with liquid baits.

Malathion bait applied to concrete floors or sacks in steer feeding barns was found to be unsatisfactory by Guthrie and Baker (1954). They concluded that poor results might be due to molasses in feeding troughs which competed as an attractant with the poisoned bait. Poisoned baits gave satisfactory results in preliminary tests in dairy barns and chicken houses where other attractants were not present.

Treatment of garbage dumps with malt baits containing 1% Shell OS 2046 (1-methoxycarbonyl-1-propen-2yl dimethyl phosphate) or Shell OS 1808 (2-carbethoxy-1-methylvinyl diethyl phosphate) or 2% malathion was effective (Keller, Wilson, and Smith, 1955). Less effective baits were 0.2% Bayer L 13/59 in sugar solution and 2% malathion in black strap molasses.

Granular baits were used by Keller and Wilson (1955) to give good control of house flies. A bait composed of 88% cornmeal, 10% sugar, and 2% malathion yielded satisfactory results with daily applications during a 4-day period. A similar bait with 1% Diazinon as the insecticide gave satisfactory control after the second daily treatment.

Keller (1955) used 1 or 2% Chlorothion (O,O dimethyl O-(3-chloro-4-nitrophenyl) thiophosphate) sugar bait to obtain satisfactory results at military installations. Chlorothion, Diazinon, and Bayer L 13/59 sugar baits gave excellent kills but no residual effects (Hansens, Granett, and O'Connor, 1955). They stated that bait results were not comparable to those of residual sprays.

Malathion, Diazinon, Pirazinon (O,O-diethyl O-3-(2-propyl-6-methyl-4-pyrimidinyl)-thiophosphate), Chlorothion, American Cyanamid 4124 (O-(2-chloro-4-nitrophenyl) O,O-dimethyl phosphorothioate), and Bayer L 13/59 sugar baits, both wet and dry, at 1% gave 90 to 100% kill in 2 hours under laboratory conditions (Johnson, Langford, and Lall, 1956). With few exceptions, flies fed on all baits promptly. Baits prepared with Bayer L 13/59 appeared to be more attractive than others. Flies were evidently repelled at initial feeding by American Cyanamid 4124, but knockdown was fast.

Keller, Wilson, and Smith (1956) used bait stations consisted of wooden tongue depressors stapled to paper coasters or pieces of screen coated with a slurry containing an insecticide, sugar, and sometimes agar, sand, or other ingredients. They reported that baits containing Bayer L 13/59 or malathion gave effective control for 28 to 98 days under conditions of fair sanitation.

Hansens (1956) describes a "plate" method of testing baits. This method consisted of exposing baits on plates or other containers in a barn and comparing effectiveness by counting flies visiting the baits or dead flies on the dishes, He placed the containers 2 feet apart, and counted 10 times each 30 minutes for a 2-hour period. Hansens also evaluated dry sugar bait grid counts in stanchions. Diazinon and Bayer L 13/59 baits gave excellent results. Dow ET-14 (0,0 dimethyl O-2,4,5-trichlorophenyl phosphorothioate) and Dow ET-15 (O-methyl O-2,4,5-trichlorophenyl phosphoramidothioate) were not quite as good. American Cyanimide 4124 and Ortho dry bait were less effective than those previously mentioned, and malathion and Fasco fly flakes were unsatisfactory. Diazinon was the superior wet bait.

Synergized pyrethrins (pyrethrum) and allethrin (dl-2-allyl-4 hydroxy-3-methyl-2-cyclopenten-1-one esters of cis and trans dl-chrysanthemummonocarboxylic acids) baits were used for the control of organophosphorus-resistant house flies (LaBrecque, Watson, and Gahan, 1958). These baits were less effective than Dipterex (Bayer L 13/59) against malathion-resistant flies. In field tests, both 0.1% pyrethrins and 0.1% allethrin were superior to Dipterex.

Laboratory and field tests were conducted with 1% Dimetilan (3-methylpyrazol-(5)-yl dimethylcarbamate) sugar baits by Smith et al. (1960). They found that the bait was effective against organophosphorus-resistant house flies in laboratory tests and in one of two field tests in dairy barns. Ribbons impregnated with a sweet syrup and Dimetilan and cords impregnated with a mixture of parathion (O,O-dimethyl O-p-nitrophenyl phosphorothioate) and Diazinon gave practical control of house flies in hog parlors but not in barns or poultry houses.

BEHAVORIAL RESISTANCE.--Behavorial resistance of flies to malathion baits was indicated in a study by Schmidt, and LaBrecque (1959). Dipterex baits showed no evidence of resistance. They concluded that the degree of avoidance demonstrated in their experiments could be one of the factors contributing to the lack of control with baits in the field, though less important than physiological resistance.

Gahan et al. (1953) discovered that DDT-resistant flies demonstrated a pattern of behavorial resistance. Concentrations of TEPP (tetraethyl pyrophosphate) above 0.5% were somewhat repellent to these flies.

VALUE OF DDVP AS A HOUSE FLY INSECTICIDE.--The value of DDVP (O,O-dimethyl 2,2-dichlorovinyl phosphate) as an insecticide was first reported in 1955 (Mattson, Spillane, and Pearce, 1955; Kilpatrick, and Schoof, 1955; Fluno, 1955). Kilpatrick and Schoof stated that preliminary studies by them demonstrated that DDVP offers considerable promise as a toxicant in baits for house fly control. They found that the effective concentration of DDVP can be reduced well below the

0.1% level commonly utilized for other organophosphorus compounds such as malathion, Bayer L 13/59, and Diazinon. Liquid baits containing 10% sugar and 0.01% DDVP gave control for 24 hours. A similar bait with 0.1% DDVP gave control for 48 hours.

Complete mortality of house flies was attained in the laboratory with sugar bait containing 0.01% DDVP (Fluno, 1955). He found that DDVP and its homologs gave greater reductions of house fly numbers than Bayer L 13/59 as sugar baits in the first 10 minutes, but control was equal after 4 hours. Schoof and Kilpatrick (1957) indicated that DDVP baits gave immediate reduction of fly populations at a dairy.

The most effective insecticides in an extensive series of fly bait tests were Dibrom (1,2-dibromo-2,2-dichloroethyl dimethyl phosphate) and DDVP (Howell, 1961). He stated that 0.25 to 0.5% of the insecticide in the bait was the most effective.

In laboratory tests on knockdown and kill with 22 organophosphorus compounds against insecticide-susceptible female house flies, the most rapidly acting and toxic compounds were DDVP, Dibrom, Diazinon, Parathion, and ronnel (O,O-dimethyl O-2,4,5-trichlorophenyl phosphorothioate) (Eddy, 1961). The flies were confined on plywood panels which had been sprayed with acetone solutions of the insecticides.

Lorber (1958) reported that DDVP used as a fumigant offers a new approach to the control of ectoparasites on animals. DDVP was the fastest acting fumigant tested against house flies with regard to knockdown (Ihndris and Sullivan, 1958).

CROSS RESISTANCE.—Forgash and Hansens (1960) studied the toxicity of insecticides to Diazinon-resistant house flies. They found a low level of increased tolerance (4 to 7 fold) with DDVP, Dipterex, Dibrom, and Phosdrin (1-methoxycarbonyl-1-propen-2-yl-dimethyl phosphate) Cross resistance to phosphates and phosphonates was less pronounced than to most phosphorothioates and some dithioates.

TOXICOLOGICAL PROPERTIES OF DDVP.--DDVP is a relatively safe insecticide to use. Radeleff and Woodward (1957) found that it was not lethal when given orally to calves or sheep. Also, 1% spray and dip were not toxic to cattle.

The acute oral toxicity of DDVP to Sherman white rats was studied by Gaines (1960). The acute oral toxicities to male and female rats were 80 and 56 mg./kg., respectively. The acute dermal toxicities to male and female rats were 107 and 75 mg./kg., respectively.

The toxicological aspects of DDVP in calves, horses, and white rats were considered by Tracy, Woodcock, and Chodroff (1960). Five horses exposed continuously to the vapors of 0.5 mg. DDVP/cu. ft. in a closed barn for 22 days displayed mild cholinesterase (ChE) depression after 7 days. They recovered to normal ChE concentration at 11 days. The plasma was within normal limits of ChE activity throughout the test period. The concentration of DDVP in the air varied between 0.24 and 1.48 $\mu$g. or 1.4 to 8.4% of the daily DDVP dose. They also demonstrated that two cows with suckling calves showed normal ChE levels while ingesting 200 p.p.m. DDVP daily. Severe depression occured at 500 p.p.m. DDVP. A single dose of 27 mg./kg. caused cholinergic collapse without recovery. The ChE levels in the calves remained normal throughout the 78-day test. Female rats nursing litters were repeatedly intoxicated by oral administration of 30 mg./kg. The litters of these rats exhibited normal ChE levels and normal weight-growth curves. Fresh liver macerates from different animals detoxified DDVP at a rate of about 500 $\mu$g. DDVP/g. liver. Tracy, Woodcock, and Chodroff concluded that DDVP is completely detoxified in the animal body and does not accumulate nor store in animal tissues. They suggest that the hazard of DDVP is related directly to the weight of acute intake.

## MATERIALS AND METHODS

TEST INSECTS.--House flies, *Musca domestica* L., were collected from the Experimental Beef Barn of Oklahoma State University in October of 1961. A culture was maintained at the Insectary of Oklahoma State University for use in laboratory tests. Chemical Specialities Manufacturer's Association (CSMA) rearing medium (Ralston Purina Company) was used. Adult flies to be used in laboratory tests were maintained on equal parts of granulated sugar and powdered dried milk solids.

Natural populations of house flies at the Experimental Beef Barn were used in field testing.

TEST LOCATIONS.--Laboratory tests were conducted in a basement room of the Entomology Field Laboratory of Oklahoma State University. The test room was 22.5 x 10.5 x 8.5 feet. The doorway of the test room was covered with a canvas flap which allowed the experimenter to go to and from the room without losing large numbers of flies. An exhaust fan (700 cu. ft./min. rating) was centrally located near the ceiling of the room to provide air movement. This prevented fumigation of flies by baits being tested.

Field experiments were conducted at the Experimental Beef Barn. Ten pens, each 40 x 27 feet, made up the greater area of the barn. These pens were occupied by beef animals during test periods. A walkway, 8 x 270 feet, was located adjacent to the pens. The experiments were conducted on this walkway.

TEST EQUIPMENT.--White, plasticized, picnic plates, 10.5 inches in diameter with 0.75-inch lips, were used in both laboratory and field tests. The lips of these plates were thought to be very effecient in retaining flies which fed on the baits distributed in them. The plates were placed approximately 1.5 feet apart and were used only once. In field tests, these plates were weighted with 1-inch iron washers which were taped to their undersides.

Two different sizes of measurers were employed to transfer uniform portions of bait from storage jars to the test plates. One-fourth teaspoon measurers were used in field testing. Approximately 1.6 g. was used per plate. Much smaller measurers were constructed from soda straws (4 mm. in diameter) and flat wooden toothpicks for use in laboratory tests. Approximately 0.03 g. was used per plate.

BAIT PREPARATION.--Several series of baits were formulated during the testing program. A technical grade of insecticide was used. It was 100% insecticidally active and contained at least 93% DDVP. Because of the unstable characteristics of DDVP, two different materials were used in various baits to stabilize the insecticide. These were Beta-naphthol and Bisphenol A (4,4'-Isopropylidenediphenol). Since granulated sugar becomes sticky upon exposure, two materials were evaluated as bait conditioners. These were Santocel C and Dicilite (expanded perlite powder).

The general mixing procedure for all baits except those prepared by Shell Chemical Company was as follows: (1) samples (0.5 pound) of a sand and sugar base with premixed stabilizers were placed in 1-pint fruit jars, (2) 2 ml. of reagent grade chloroform with the appropriate aliquot of DDVP were mixed into the base materials with a glass stirring rod, (3) the jars were hand rotated until the ingredients had been throughly mixed, (4) 18 to 24 hours later the conditioners were added to the appropriate jars, (5) baits were exposed for approximately 30 minutes, and (6) the jars were hand rotated to provide through mixing of the ingredients.

TEST PROCEDURES.--Baits for both field and laboratory experiments were distributed on plates in the Entomology Field Laboratory. The positions of the plates in the test area were determined at random. The plates were stacked in accordance with their respective positions and transferred to the test area in this fashion. These preliminary procedures enhanced the rapidity of plate distribution in field tests. This should have reduced the experimental error. This factor was no problem in laboratory experiments. Plates were placed in their proper locations before laboratory cultures of flies were released.

MEASUREMENTS.--The method of measurement used in field experimentation was that of determining the volume of dead flies in each plate. This was done because the numbers of flies were often too numerous for visual counting. Several pieces of equipment were employed to accomplish this task. These were 100 ml. graduated cylinders, 10-inch (diameter) funnels, and 1.5-inch (width) paint brushes for manipulation of dead flies. These measurements were taken approximately 5 hours after application in all field tests.

The populations of flies used in the laboratory experiments were such that the numbers of dead flies per plate could be accurately counted. A mechanical hand counter was used for this purpose. Four-hour counts were made in laboratory experimentation.

STATISTICAL PROCEDURES.--All of the experiments were designed as randomized complete-blocks. The essence of this design is that the experimental material is divided into groups, each of which constitutes a single trial or replication. At all stages of the experiments, the object was to keep the experimental errors within each group as small as practicable. Treatment positions were assigned within each group at random. A separate randomization was made for each replication.

Most of the study involved the use of factorial experiments. With this type of arrangement, the effects of a number of different factors were investigated simultaneously. The treatments consisted of all combinations that could be formed from the types or levels of each factor. The objectives of the factorial experiments were as follows: (1) to determine the effects of each of a number of factors over a specified range, (2) to investigate the interactions among the effects of several factors, and (3) to lead to recommendations that could apply over a wide range of conditions.

The data were first recorded in bound field notebooks. They were next transferred to standard note paper and all measurements in ml. were transformed to fly numbers. This was done by multiplying ml. by flies/ml. Ten samples of 5, 10, and 20 ml. of flies were counted to determine flies/ml. The average number of flies/ml. was 10.45 for 1962 and 13.7 for 1961. The data were then punched on International Business Machine (IBM) card to facilitate analyses on a high speed computer (IBM model 650).

PRELIMINARY EXPERIMENTS WITH DDVP AND DIBROM BAITS.--Tests were made with many promising candidate insecticides for use in dry sugar fly baits during the summer of 1960 at the Oklahoma Experiment Station (Howell, 1961). He reported that two organic phosphate insecticides consistently gave the highest and most rapid kills. These were DDVP and Dibrom.

A large series of treatments (21) were field tested during the summers of 1961 and 1962 in order to gain further information on the performance of these two insecticides and other bait constituents. These baits were prepared by Shell Chemical Company and shipped to Oklahoma State University during June of 1961. The treatments are given in table 1. The 1961 experiment was conducted 17 days over a 60-day period. Four replications were made per day. The same baits were tested on 4 days over an 82-day period during the summer of 1962. In the 1962 test, three replications were made per day.

3 X 3 X 5 FACTORIAL.--This experiment was designed to study the effects of three factors under field conditions. The factors were insecticide stabilizer, bait conditioner, and DDVP insecticide. The stabilizers were 0.33% Betanaphthol, 0.5% Bisphenol A, and no-stabilizer. The conditioners were 0.5% Santocel C, 1% Dicilite, and no-conditioner. The concentrations of DDVP were 1, 0.1, 0.01, 0.001, and 0%. The treatments all contained from 67.5 to 70% Ottawa sand and 30% granulated sugar. A 3 x 3 x 5 factorial arrangement of treatments was used. The test was conducted on 12 days throughout an 80-day period. Two replications were made per day.

3 X 4 FACTORIAL.--A wide range of DDVP concentrations and stabilizers were considered in this field experiment. Stabilizers were 0.33% Betanaphthol, 0.5% Bisphenol A, and no-stabilizer. Concentrations

Table 1.--Bait formulations (% by wt.) for preliminary experiments with DDVP and Dibrom, 1961 and 1962.

| Bait No. | DDVP | Ottawa Sand | Granulated Sugar | Dicilite | Polyvinyl-chloride | Beta-naph. | Phenol | Dibrom |
|---|---|---|---|---|---|---|---|---|
| 1 | 0.50 | 69.25 | 30.0 | - | - | - | 0.25 | - |
| 2 | 0.50 | 68.25 | 30.0 | 1.0 | - | - | 0.25 | - |
| 3 | 0.50 | 68.50 | 30.0 | - | - | 1.0 | - | - |
| 4 | 0.10 | 66.90 | 30.0 | - | 2.0 | 1.0 | - | - |
| 5 | 0.25 | 66.75 | 30.0 | - | 2.0 | 1.0 | - | - |
| 6 | 0.10 | 69.90 | 30.0 | - | - | - | - | - |
| 7 | 0.05 | 67.70 | 30.0 | - | 2.0 | - | 0.25 | - |
| 8 | 0.51 | 66.49 | 30.0 | - | 2.0 | 1.0 | - | - |
| 9 | 0.05 | 66.95 | 30.0 | - | 2.0 | 1.0 | - | - |
| 10 | 0.50 | 69.50 | 30.0 | - | - | - | - | - |
| 11 | 0.10 | 67.65 | 30.0 | - | 2.0 | - | 0.25 | - |
| 12 | 0.25 | 67.50 | 30.0 | - | 2.0 | - | 0.25 | - |
| 13 | 0.05 | 69.95 | 30.0 | - | - | - | - | - |
| 14 | 0.25 | 69.75 | 30.0 | - | - | - | - | - |
| 15 | - | 66.50 | 30.0 | - | 2.0 | 1.0 | - | 0.50 |
| 16 | 0.50 | 67.25 | 30.0 | - | 2.0 | - | 0.25 | - |
| 17 | - | 67.50 | 30.0 | - | 2.0 | - | - | 0.50 |
| 18 | - | 66.75 | 30.0 | - | 2.0 | - | 0.25 | 0.50 |
| 19 | 0.50 | - | 99.5 | - | - | - | - | - |
| 20 | - | - | 100.0 | - | - | - | - | - |
| 21 | - | - | - | - | - | - | - | - |

of DDVP were 10, 1, 0.1, and 0%. All of the treatments contained from 59 to 69.5% Ottawa sand, 30% granulated sugar, and 0.5% Santocel C. A 3 x 4 factorial arrangement of treatments was considered. The test was conducted on three consecutive days. Five replications were made per day.

2 X 10 FACTORIAL.--This experiment was designed to detect possible differences within a rather narrow range of DDVP concentrations and the effects of two stabilizers. Stabilizers were 0.33% Betanaphthol and no-stabilizer. Concentrations of DDVP were 2, 1.8, 1.6, 1.4, 1.2, 1, 0.8, 0.6, 0.4, and 0.2%. All of the treatments contained from 67.17 to 69.3% Ottawa sand, 30% granulated sugar, and 0.5% Santocel C. The treatments consisted of a 2 x 10 factorial arrangement. This field test was conducted on 3 consecutive days. Three replications were made per day.

COLORED BAITS.--It has been suggested by various workers (Freeborn and Berry, 1935; Harsham, 1946; Waterhouse, 1948; and Howell, 1961) that color is an important factor in the attraction of house flies to treated surfaces or baits. Therefore, field experiments were conducted to study the effect of color in sugar baits. Nine colors were compared. These were white, red, green, orange, yellow, blue, violet, brown, and black. Nine of the baits contained 99% granulated sugar, 0.5% DDVP, and 0.5% powdered chalk. A check bait contained 99.5% granulated sugar and 0.5% DDVP. Two tests were conducted on consecutive days. Seven replications were made in each test. The first test was made with 9-inch, white paper plates. The second test was made with 9-inch, white paper plates with 6.5-inch (diameter) circles of black construction paper glued in the center.

BLACK AND WHITE CONTRAST.--An experiment was designed to study black and white color contrast of three factors. The factors were background, paper plate, and sugar bait. The experimental design was a split-plot with a 2 x 2 factorial arrangement of sub-plot treatments. The main plot treatments were black and white backgrounds. Background was provided by 3 x 5 feet sections of crepe paper. The sub-plot treatments consisted of a factorial arrangement of black and white paper plates and black and white sugar baits. The plates were colored by aerosol bomb application of enamel paint. The baits contained 99% granulated sugar, 0.5% DDVP, and 0.5% powdered chalk. This experiment was conducted in the laboratory test room on 2 consecutive days with seven replications per day.

SHELF LIFE.--The effectiveness of 1% DDVP baits used periodically throughout the fly season and newly mixed baits was studied in the laboratory to determine shelf life over this period. The stored baits were used in a previous test (3 x 3 x 5 factorial) and were formulated June 25, 1962. The fresh baits were prepared October 16, 1962. Therefore, the stored baits were 113 days older than the freshly prepared baits. The experiment was a 3 x 2 factorial. The factors were conditioner and length of storage. The three conditioners were 0.5% Santocel C, 1% Dicilite, and no-conditioner. Lengths of storage were 113 to 116 and 0 to 3 days. All the baits contained from 67.67 to 68.67% Ottawa sand, 30% granulated sugar, 1% DDVP, and 0.33% Beta-naphthol. The test was conducted on 4 consecutive days with six replications per day.

BAIT EXPOSURE.--A study was designed to collect information on the effectiveness of DDVP sugar bait with regard to exposure time. Three tablespoon measures of bait were exposed in the 10.5-inch paper plates. The baits were used in the 2 x 10 factorial previously described. These exposed portions were used at various intervals (4, 5, 12, and 13 days) to make up $\frac{1}{2}$ of the treatments in the test. The other $\frac{1}{2}$ of the treatments were baits taken from the same mixtures which were used in the 2 x 10 factorial. These baits were designated as fresh baits. A 2 x 2 x 5 factorial arrangement of treatments was considered. The factors were stabilizer, exposure time, and DDVP concentration. The stabilizers were 0.33% Betanaphthol and no-stabilizer. The exposure times were considered as 0 for fresh baits and 4, 5, 12, or 13 days for exposed baits. The concentrations of DDVP were 2, 1.6, 1.2, 0.8, and 0.4%. Four field tests were made at the day intervals indicated above. Three replications were made per day.

## RESULTS

PRELIMINARY EXPERIMENTS WITH DDVP AND DIBROM.--House fly, *Musca domestica* L., mean kills for the various bait treatments considered in 1961 and 1962 are given in order of decreasing effectiveness in table 2 of appendix A. The treatment numbers in this table correspond to the formulations given in table 1 of the materials and methods section. The analyses of variance for the 1961 and 1962 experiments are presented in tables 13 and 14, respectively, of appendix B. The F values for baits in both experiments are significant at the 1% level of probability; however, the 1961 experiment includes much more replication than the one for 1962. A valid comparison of the efficiencies of these experiments can be made with the coeffecients of variation. The coeffecients of variation for 1961 and 1962 were 113.8 and 38.2%, respectively. This marked increase in efficiency in 1962 indicates that considerable progress was made in test procedures and design.

The effect of DDVP concentration in various baits of the 1961 experiment is illustrated in figure 1 of appendix C. A linear response was noted by the corresponding increases in mean fly kill with each increase in DDVP concentration.

Figure 2 demonstrates the interaction between stabilizers and DDVP concentrations with polyvinylchloride baits. With 0.5% DDVP, 0.25% phenol was the better stabilizer. However, 1% Betanaphthol gave higher kills at DDVP concentrations of 0.1 and 0.05%.

The effect of time with regard to several 0.5% DDVP baits in the 1961 experiment is considered in figure 3. Each 2-week point for each bait is the mean of 16 replications except point 4. Twenty replications are considered at this point. Percent of the 2-week mean was computed by dividing the bait means by the grand mean for each 2-week period and multiplying by 100. The upward trend of the Dicilite and phenol combination was noted. The opposite of this was indicated for polyvinylchloride baits. A slight upward trend was noted for bait no. 10 (no-additives); however, it was markedly lower than the other 0.5% baits for the first three 2-week periods.

The performance of the three Dibrom baits during the 1961 experiment is given in figure 4. The percentages given here were computed in the manner of the preceeding figure. Little difference was noted between the Betanaphthol and phenol baits over the four 2-week periods. The line for bait no. 17 (no-additives) was similar to the line for DDVP bait no. 10 in figure 3. This suggested that the response for the two insecticides was about the same when no stabilizers or conditioners were present.

3 X 3 X 5 FACTORIAL.--Table 3 gives the stabilizers x conditioners x concentrations means for the 3 x 3 x 5 factorial. The analysis of variance is presented in table 15. F values for the main effects (stabilizers, conditioners, and concentrations) were significant at the 1% level. Also, a majority of the interaction F values were significant at either the 1 or 5% level of probability. The coeffecient of variation for this experiment was 39%. This was very close to 38.2% for the preliminary test with DDVP and Dibrom in 1962.

The effect of DDVP concentration in the 3 x 3 x 5 factorial is illustrated in figure 5. This effect was somewhat similar to that in figure 1 for the preliminary experiment with DDVP and Dibrom, 1961. Little difference was seen in the mean fly kill for 1 and 0.1% baits. These were decidedly superior in performance as compared to the lower concentrations.

The performance of the various DDVP concentrations over the four 3-week periods of the test are given in figure 6. Some interaction was present as would be expected by the highly significant F value of 3.18 for days x concentrations in the analysis of variance (table 15). It was noted that the 1 and 0.1% concentration lines indicated an upward trend in their performance. A downward trend was suggested by the lower concentration lines.

The effect of the various stabilizers over the four 3-week periods is illustrated in figure 7. The main effect mean for no-stabilizer (134) was just slightly higher than for Betanaphthol (133.36). However, the lines for these two factors indicated an interaction with time. Also, the highly significant F value of 5.61 (table 15) for days x stabilizers indicated an interaction effect. The Betanaphthol formulations were lower for the first two 3-week periods and higher for the last two as compared to no-stabilizer. The Bisphenol A stabilizer consistently gave the lowest performance throughout the test period.

Stabilizers x conditioners is considered in figure 8. Very little interaction between these factors was present as suggested by the small F value of 1.45 (table 15) for this effect.

Stabilizers x concentrations interaction was indicated by the highly significant F value of 5.08 in table 15. This was further considered in figure 9. It was noted that Betanaphthol and no-stabilizer showed interaction between the 1 and 0.1% DDVP concentrations. Betanaphthol gave better results at 1% and no-stabilizer at 0.1%.

The main effect mean fly kills for the various conditioners were computed from the appropriate means in table 3. These were 125.97, 106.25, and 87.24 for Santocel C, Dicilite, and no-conditioner, respectively.

3 X 4 FACTORIAL.--The analysis of variance for this experiment is presented in table 16. The F values for the main effects of stabilizers and concentrations were both significant at the 1% level of probability. Stabilizers x concentrations was significant at the 5% level.

The means for this experiment are presented in table 4. The no-stabilizer mean was higher than the Betanaphthol mean which was better than Bisphenol A. The effect of DDVP concentration is illustrated in figure 10. The 10% concentration was decidedly lower in mean fly kill than the others. The fact that it was lower than 0% suggested that 10% was unattractive to house flies. Also, the mean of 179.07 for 0% DDVP was much higher in relation to the grand mean than for 0% in other experiments in this report.

2 X 10 FACTORIAL.--The analysis of variance for this experiment is given in table 17. The F value of 15.74 for stabilizers was highly significant. In table 5, the mean for no-stabilizer over all DDVP concentrations was 341.98 as compared to 239.56 for Betanaphthol. The concentration means in table 5 showed only small differences, and

certainly no general trend was detected. The F values of 1.06 for concentrations and 1.82 for stabilizers x concentrations were not significant at the 5% level.

COLORED BAITS.--The means for the various colored baits on both white and black paper plates are given in table 6. The analyses of variance for these experiments can be found in tables 18 and 19, respectively. Only small differences were noted in the means for colored baits on either white or black plates. The F value of 1.81 for colored baits on black plates was much higher than 0.58 for white plates. Neither of these was significant at the 5% level, however.

BLACK AND WHITE CONTRAST.--The analysis of variance for the black and white contrast study is in table 20. The backgrounds x plates x baits means are presented in table 7. The main effect means for backgrounds can also be noted in this table. The main effect means for plates and baits can be computed from the appropriate means in table 7. The F values in the analysis of variance table for backgrounds and baits were significant at the 1 and 5% levels of probability, respectively. This value was not significant at the 5% level for plates.

Plates x baits was considered in figure 11. Black bait was superior to white bait for both black and white plates, but the lines came much closer together for black plates than white. The plates x baits interaction F value was not significant at the 5% level.

The mean fly kill lines for backgrounds x baits were plotted in figure 12. Black bait gave higher kills for both white and black plates. Very little, if any, interaction was indicated for these two factors. The small F value of 0.66 from table 20 did not suggest any interaction in this case.

Figure 13 illustrates the mean response of backgrounds x plates. The lines were much closer together here than in the other two illustrations for this experiment. Less interaction was detected for these factors than for the other two combinations. Black colored plates yielded the highest kills for both white and black backgrounds.

SHELF LIFE.--The analysis of variance for the shelf life study is presented in table 21. The only significant F value in the analysis was for days x conditioners. This interaction is illustrated in figure 14. Dicilite baits went from high to low in performance. No-conditioner baits were just the opposite. The response to Santocel C formulations was most consistent for all 4 days.

The main effect means for stored and fresh bait were computed from the appropriate means in table 8. These were 19.96 and 28.93 for stored and fresh baits, respectively. The days x conditioners x storages means can be found in table 8.

BAIT EXPOSURE.--Stabilizers x concentrations x exposures means for experiments 1, 2, 3, and 4 of this study are given in tables 9, 10, 11, and 12, respectively. Marked differences occurred between exposed and fresh baits except in experiment 3 (12th day of exposure). The stabilizers x exposures means can be computed from the appropriate means from the tables previously mentioned. It was interesting to note that Betanaphthol gave better results than no-stabilizer for exposed baits in each experiment except no. 3. Some DDVP concentration differences were detected in experiments 2 and 4. The analyses of variance for these experiments are presented in tables 22, 23, 24, and 25, respectively.

## DISCUSSION

The results in the foregoing section have been presented with respect to the individual experiments or types of experiments. This section of the report is intended to integrate the effects of the factors from all experiments in the study. Also, hypotheses are given to help explain the results of the experimentation.

CONCENTRATION OF INSECTICIDE.--The effect of DDVP concentration up to 1% in fly baits approached linearity in the various experiments. Baits at 10% in the 3 x 4 factorial did not follow this pattern, however. These baits indicated that DDVP was repellent and/or deterrent (Dethier, Browne, and Smith, 1960) in its action and/or destroys the feeding qualities of granulated sugar. Baits at 10% seemed to have an effect with regard to other treatments in this experiment. The mean fly kills for 0% DDVP were unusually high in comparison with other experiments in this study. Several hypotheses could be used to explain this effect. Perhaps house flies transferred very minute portions of the 10% baits to plates containing 0% DDVP. This would kill flies feeding in these plates. Support is added to this hypothesis by the demonstration of transfer of bait material from container to container by house flies (Smith, 1962). Another explanation would be that 0% baits would elicit a greater feeding stimulus for flies receiving lethal doses from 10% baits.

No differences could be detected among a rather narrow range of concentrations in the 2 x 10 factorial. The lowest concentration of 0.2% seemed to be fairly close to the minimum required for maximum performance over a 3-day period. The highest concentration of 2% was not similar to 10% (3 x 4 factorial) in its action. If differences existed among these concentrations, they were not large enough to be detected by the precision demonstrated in this experiment.

STABILIZERS.--Preliminary experiments with DDVP and Dibrom suggested that insecticide stabilizers were of definite value as additives to house fly baits. Phenol at 0.25% was superior to 1% Betanaphthol for high concentrations of DDVP. The reverse of this was true for low DDVP concentrations. This concentration of phenol may have been adequate for high DDVP concentrations but not for low concentrations. The higher concentration of Betanaphthol may have altered slightly the attractant and/or feeding stimulant qualities of the sugar baits.

Bisphenol A was much inferior to Betanaphthol and no-stabilizer in experiments which compared the three. The results from these experiments strongly suggested that this concentration of Bisphenol A was repellent and/or deterrent in various fly bait formulations. The interaction for no-stabilizer and Betanaphthol between 1 and 0.1% DDVP concentrations is very difficult to explain. It is possible that 0.33% Betanaphthol acted somewhat as a deterrent. This would yield lower kills for low concentrations of DDVP. Less feeding would be necessary for 1% DDVP which would tend to offset the effect of Betanaphthol.

CONDITIONERS.--All of the experiments indicated that bait conditioners were of definite value in house fly bait formulations. In preliminary experiments, 1% Dicilite was superior to 2% polyvinylchloride. Other experiments showed that 0.5% Santocel C was higher in mean fly kill than 1% Dicilite.

The effect of conditioners over a 4-day period presented and interesting and somewhat puzzling picture. It is possible that this interaction may be explained by moisture content of the baits. No-conditioner baits may have increased in moisture content as the experiment progressed which could have increased the attractiveness of these formulations. Added moisture did enhance the attractiveness of baits in experimentation by other workers (Smith, 1962).

COLOR.--Field experiments with baits colored with 0.5% powdered chalk did not demonstrate any statistical significant differences among them. A laboratory experiment indicated significant differences between black and white baits and backgrounds, however. A factor that should be mentioned is that the laboratory test room was lighted with fluroscent tubes. Natural lighting was present in field experiments. The results of the laboratory study suggested a relationship between color effect and house fly preference.

SHELF LIFE.--DDVP baits at 1% stored in 1-pint fruit jars and used every 3 weeks over a 113-day period were compared to fresh baits of the same formulation and found to be only 69% as effective. It is difficult to explain why the Santocel C stored formulation was superior in performance to its fresh counterpart for the whole experiment. The means in table 8 of appendix A showed that Santocel C stored baits were markedly superior for only the first 2 days of the experiment. Fresh baits were considerably better for the last 2 days.

A similar pattern was noted for Dicilite but in a reduced degree. Perhaps the performance of these baits depended more on moisture content or ther physical factors than quantity of DDVP in the formulations.

BAIT EXPOSURE.--Marked differences occurred between exposed and fresh baits in three of four experiments. No difference could be demonstrated on the 12th day of exposure. The means in table 11 of this experiment suggested that Betanaphthol exposed baits performed much better than no-stabilizer baits. This difference was not noted in the other experiments of this study. The combined results of the exposure experiments indicated that exposed baits were 66.5% as effective as fresh baits. In general, the addition of Betanaphthol did not increase the performance of exposed baits by any marked degree.

EXPERIMENTAL DESIGN.--The advantages of the randomized complete-block design were realized in the various experiments in this report. The balance and unlimited replication features of this design simplified experimental procedures and data analyses to a great extent.

The factorial arrangements of treatments were particularly useful. The factors were not independent in most cases. Because of this, a portion of the information presented could not have been accumulated with the single-factor approach. This information was the interactions of the types and levels of the factors considered.

## SUMMARY AND CONCLUSIONS

Thirteen experiments were conducted to determine the sensory perception of various bait factors by the house, Musca domestica L. Laboratory and field tests were conducted at the Entomology Field Laboratory and the Experimental Beef Barn, respectively, of Oklahoma State University. All of the experiments were designed as randomized complete-blocks. Most of these included the use of a factorial arrangement of treatments. The data were punched on International Business Machine (IBM) cards to facilitate analyses on a high speed computer (IBM model 650).

Preliminary experiments with DDVP and Dibrom during the summers of 1961 and 1962 indicated that a majority of baits ranging in concentration from 0.05 to 0.5% remained effective for at least 15 months. Several of these baits yielded a linear response with regard to fly kill and DDVP concentration. Various baits also showed that 0.25% phenol was superior to 1% Betanaphthol as an insecticide stabilizer at DDVP concentrations of 0.5 and 0.25%, but the reverse was true at concentrations of 0.1 and 0.05%. A bait containing 0.25% phenol as the stabilizer and 1% Dicilite as the bait conditioner increased in its performance during the summer of 1961 as compared to other 0.5% DDVP bait formulations. The effectiveness of two 0.5% Dibrom baits was enhanced by the addition of stabilizers and polyvinylchloride conditioner.

The coeffecients of variation for the same series of treatments for summers of 1961 and 1962 were 113.8 and 38.2%, respectively. These values showed that an increase was made in test efficiency for the latter summer.

An extensive experiment involving three stabilizers, three conditioners, and five concentrations of DDVP was conducted over an 80-day period. The mean fly kills for DDVP concentrations ranging from 0 to 1% indicated linearity. DDVP formulations at 1% gave only slightly higher kills than those at 0.1%. A concentrations x time study revealed that 1 and 0.1% baits progressed in performance over the test period as contrasted to degression by lower concentrations.

A 0.5% Bisphenol A stabilizer was much lower in mean fly kill than 0.33% Betanaphthol and no-stabilizer. The latter two were similar in mean fly kills. However, the stabilizers x time investigation revealed an upward trend in the performance of Betanaphthol baits in contrast to a downward trend by no-stabilizer.

Baits containing 0.5% Santocel C were superior to those with 1% Dicilite which were better than no-conditioner formulations.

A repellency effect of high concentrations of DDVP was noted when 10% baits were found to be markedly inferior in performance to concentrations of 1, 0.1, and 0%.

No substantial differences were detected in a series of baits ranging in concentration from 0.2 to 2% DDVP.

No differences could be demonstrated among 10 colored baits in field experiments. These baits were evaluated on both black and white plates. A laboratory study was made on black and white contrast of three factors. These were backgrounds, plates, and baits. Black gave the highest mean fly kills for all factors; however, the difference in plates was not significant at the 5% level of probability. Some interaction was noted for the plates x baits study, but it was not significant at the 5% level. No interactions were detected for either backgrounds x baits or backgrounds x plates.

Mean fly kills of 19.96 and 28.93 were yielded by stored and fresh baits, respectively, in a shelf life study. A study of the days x conditioners interaction suggested that 1% Dicilite formulations became progressively less effective over the 4 consecutive days of the experiment. No-conditioner baits increased in performance over the same period. Santocel C baits were the most consistent in performance.

Fresh baits were more effective than exposed baits in three of four experiments. It was noted that 0.33% Betanaphthol stabilized baits gave better results than no-stabilizer for exposed baits in three of four tests.

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APPENDIXES

APPENDIX A

Table 2.--Number[a] and rank of treatment means for preliminary experiments with DDVP and Dibrom, 1961 and 1962.

| 1961 | | 1962 | |
|---|---|---|---|
| No. | Mean | No. | Mean |
| 15 | 289.96 | 1 | 405.75 |
| 1 | 283.21 | 3 | 403.33 |
| 18 | 262.99 | 2 | 391.67 |
| 2 | 249.04 | 18 | 389.42 |
| 16 | 247.31 | 8 | 372.83 |
| 3 | 217.97 | 10 | 371.00 |
| 8 | 212.49 | 17 | 366.83 |
| 19 | 189.29 | 16 | 353.42 |
| 17 | 175.97 | 5 | 352.17 |
| 12 | 171.07 | 12 | 348.58 |
| 5 | 168.47 | 15 | 337.00 |
| 4 | 154.32 | 9 | 322.17 |
| 10 | 137.14 | 4 | 320.42 |
| 6 | 136.18 | 19 | 317.83 |
| 9 | 121.60 | 11 | 272.08 |
| 14 | 120.78 | 14 | 254.17 |
| 11 | 113.88 | 13 | 225.42 |
| 13 | 74.81 | 6 | 167.67 |
| 7 | 47.15 | 7 | 87.58 |
| 20 | 20.76 | 20 | 61.67 |
| 21 | 13.85 | 21 | 25.92 |
| Mean | 162.30 | | 292.71 |

[a]Formulations given in table 1.

Table 3.--Stabilizers x conditioners x concentrations means for 3 x 3 x 5 factorial.

| Stabilizer | Conditioner | Concentration 1% | 0.1% | 0.01% | 0.001% | 0% | Mean |
|---|---|---|---|---|---|---|---|
| Beta-naphthol | Santocel C | 284.1 | 195.1 | 106.2 | 104.6 | 86.8 | 155.4 |
| | Dicilite | 302.5 | 218.6 | 100.5 | 14.3 | 10.7 | 129.3 |
| | No-cond. | 229.3 | 219.5 | 95.8 | 16.1 | 16.3 | 115.4 |
| Bis-phenol A | Santocel C | 134.6 | 86.6 | 36.2 | 13.6 | 3.7 | 54.9 |
| | Dicilite | 140.0 | 54.6 | 43.0 | 7.8 | 13.9 | 51.9 |
| | No-cond. | 83.9 | 97.4 | 33.7 | 18.8 | 8.3 | 48.4 |
| No-stabilizer | Santocel C | 198.0 | 267.5 | 251.4 | 41.7 | 79.5 | 167.6 |
| | Dicilite | 177.9 | 234.3 | 54.9 | 172.5 | 42.7 | 136.5 |
| | No-cond. | 135.7 | 231.8 | 66.4 | 24.8 | 30.7 | 97.9 |
| Mean | | 187.4 | 178.4 | 87.6 | 46.0 | 32.5 | 106.4 |

Table 4.--Stabilizers x concentrations means for 3 x 4 factorial.

| Stabilizer | Concentration 10% | 1% | 0.1% | 0% | Mean |
|---|---|---|---|---|---|
| Betanaphthol | 45.53 | 160.87 | 252.93 | 213.13 | 168.12 |
| Bisphenol A | 71.00 | 192.40 | 172.73 | 102.60 | 134.68 |
| No-stabilizer | 66.87 | 287.00 | 259.07 | 221.47 | 208.60 |
| Mean | 61.13 | 213.42 | 228.24 | 179.07 | 170.47 |

Table 5.--Stabilizers x concentrations means for 2 x 10 factorial.

| Concentration (%) | Stabilizer | | Mean |
|---|---|---|---|
| | Betanaphthol | No-stabilizer | |
| 2.0 | 370.44 | 298.33 | 334.39 |
| 1.8 | 216.00 | 304.11 | 260.06 |
| 1.6 | 203.11 | 371.56 | 287.33 |
| 1.4 | 218.33 | 351.89 | 285.11 |
| 1.2 | 159.00 | 484.11 | 321.56 |
| 1.0 | 211.33 | 376.33 | 293.83 |
| 0.8 | 185.78 | 283.44 | 234.61 |
| 0.6 | 213.67 | 282.33 | 248.00 |
| 0.4 | 230.00 | 311.11 | 270.56 |
| 0.2 | 387.89 | 356.56 | 372.22 |
| Mean | 239.56 | 341.98 | 290.77 |

Table 6.--Ranked means for colored baits on white and black plates.

| White Plates | | Black Plates | |
|---|---|---|---|
| Color | Mean | Color | Mean |
| Red | 429.86 | Check (white)[a] | 591.29 |
| Black | 428.43 | Violet | 540.57 |
| Orange | 403.14 | Brown | 516.57 |
| Violet | 401.57 | Blue | 476.29 |
| Blue | 398.57 | Yellow | 465.71 |
| Green | 395.57 | Black | 464.29 |
| Yellow | 394.14 | White | 455.43 |
| Brown | 368.86 | Green | 449.29 |
| White | 349.43 | Orange | 447.71 |
| Check (white)[a] | 309.14 | Red | 409.14 |
| Mean | 387.87 | Mean | 481.63 |

[a]No powdered chalk added.

Table 7.--Backgrounds x plates x baits means for black and white contrast study.

| Plate | Bait | Background: White | Background: Black | Mean |
|---|---|---|---|---|
| White | White | 3.86 | 15.43 | 9.64 |
| | Black | 30.00 | 75.57 | 52.79 |
| Black | White | 22.36 | 42.57 | 32.46 |
| | Black | 42.00 | 58.86 | 50.43 |
| Mean | | 24.55 | 48.11 | 36.33 |

Table 8.--Days x conditioners x storages means for shelf life study.

| Conditioner | Storage | Days: 1 | 2 | 3 | 4 | Mean |
|---|---|---|---|---|---|---|
| Santocel C | Stored | 42.50 | 47.67 | 11.33 | 5.83 | 26.83 |
| | Fresh | 25.00 | 25.33 | 27.50 | 18.33 | 24.04 |
| Dicilite | Stored | 27.17 | 42.50 | 8.67 | 3.50 | 20.46 |
| | Fresh | 46.17 | 82.17 | 25.17 | 7.67 | 40.29 |
| No-conditioner | Stored | 4.67 | 9.33 | 7.83 | 28.50 | 12.58 |
| | Fresh | 4.33 | 31.33 | 18.83 | 35.33 | 22.46 |
| Mean | | 24.97 | 39.72 | 16.56 | 16.53 | 24.44 |

Table 9.--Stabilizers x concentrations x exposures means for experiment 1 of exposure study.

| Stabilizer | Concentration (%) | Exposure: Exposed | Exposure: Fresh | Mean |
|---|---|---|---|---|
| Betanaphthol | 2.0 | 104.67 | 132.00 | 118.33 |
| | 1.6 | 101.33 | 209.33 | 155.33 |
| | 1.2 | 73.00 | 167.00 | 120.00 |
| | 0.8 | 73.00 | 303.00 | 188.00 |
| | 0.4 | 76.67 | 164.67 | 120.67 |
| No-stabilizer | 2.0 | 111.33 | 55.67 | 83.50 |
| | 1.6 | 7.00 | 101.33 | 54.17 |
| | 1.2 | 49.00 | 266.33 | 157.67 |
| | 0.8 | 38.67 | 171.67 | 105.17 |
| | 0.4 | 108.00 | 209.00 | 158.50 |
| Mean | | 74.27 | 178.00 | 126.13 |

Table 10.--Stabilizers x concentrations x exposures means for experiment 2 of exposure study.

| Stabilizer | Concentration (%) | Exposure: Exposed | Exposure: Fresh | Mean |
|---|---|---|---|---|
| Betanaphthol | 2.0 | 94.00 | 90.67 | 92.33 |
| | 1.6 | 111.33 | 261.33 | 186.33 |
| | 1.2 | 90.67 | 237.33 | 164.00 |
| | 0.8 | 209.33 | 383.33 | 296.33 |
| | 0.4 | 383.33 | 289.33 | 336.33 |
| No-stabilizer | 2.0 | 62.67 | 111.67 | 87.17 |
| | 1.6 | 83.33 | 212.33 | 147.83 |
| | 1.2 | 184.67 | 345.00 | 264.83 |
| | 0.8 | 177.67 | 240.33 | 209.00 |
| | 0.4 | 160.00 | 125.67 | 142.83 |
| Mean | | 155.70 | 229.70 | 192.70 |

Table 11.--Stabilizers x concentrations x exposures means for experiment 3 of exposure study.

| Stabilizer | Concentration (%) | Exposure: Exposed | Exposure: Fresh | Mean |
|---|---|---|---|---|
| Betanaphthol | 2.0 | 146.00 | 163.67 | 154.83 |
| | 1.6 | 118.67 | 167.33 | 143.00 |
| | 1.2 | 163.67 | 125.67 | 144.67 |
| | 0.8 | 191.67 | 118.33 | 155.00 |
| | 0.4 | 219.33 | 59.33 | 139.33 |
| No-stabilizer | 2.0 | 129.00 | 70.00 | 99.50 |
| | 1.6 | 170.67 | 205.67 | 188.17 |
| | 1.2 | 156.67 | 160.00 | 158.33 |
| | 0.8 | 128.67 | 237.00 | 182.83 |
| | 0.4 | 73.33 | 205.67 | 139.50 |
| Mean | | 149.77 | 151.27 | 150.52 |

Table 12.--Stabilizers x concentrations x exposures means for experiment 4 of exposure study.

| Stabilizer | Concentration (%) | Exposure: Exposed | Exposure: Fresh | Mean |
|---|---|---|---|---|
| Betanaphthol | 2.0 | 299.33 | 202.00 | 250.67 |
| | 1.6 | 69.33 | 160.33 | 114.83 |
| | 1.2 | 38.33 | 149.67 | 94.00 |
| | 0.8 | 149.67 | 111.67 | 130.67 |
| | 0.4 | 27.00 | 55.67 | 41.33 |
| No-stabilizer | 2.0 | 195.00 | 125.33 | 160.17 |
| | 1.6 | 67.67 | 184.67 | 126.17 |
| | 1.2 | 31.33 | 216.33 | 123.83 |
| | 0.8 | 17.67 | 240.33 | 129.00 |
| | 0.4 | 111.33 | 188.00 | 149.67 |
| Mean | | 100.67 | 163.40 | 132.03 |

# APPENDIX B

Table 13.—Analysis of variance for preliminary experiment with DDVP and Dibrom, 1961.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Baits | 20 | 450,629.60 | 13.21 |
| Days x Baits | 320 | 37,106.04 | 1.09 |
| Error | 1020 | 34,123.18 | |

Table 14.—Analysis of variance for preliminary experiment with DDVP and Dibrom, 1962.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Baits | 20 | 159,291.50 | 12.74 |
| Days x Baits | 60 | 52,326.30 | 4.19 |
| Error | 120 | 12,501.60 | |

Table 15.—Analysis of variance for 3 x 3 x 5 factorial

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 2 | 805,800.00 | 37.78 |
| Days x Sta. | 22 | 119,625.40 | 5.61 |
| Conditioners | 2 | 135,010.00 | 6.33 |
| Sta. x Cond. | 4 | 30,980.00 | 1.45 |
| Days x Cond. | 22 | 35,703.60 | 1.67 |
| Days x Sta. x Cond. | 44 | 18,028.20 | 0.85 |
| Concentrations | 4 | 1,144,330.00 | 33.65 |
| Sta. x Conc. | 8 | 108,315.00 | 5.08 |
| Cond. x Conc. | 8 | 30,092.60 | 1.41 |
| Sta. x Cond. x Conc. | 16 | 45,582.60 | 2.14 |
| Days x Conc. | 44 | 67,798.60 | 3.18 |
| Days x Sta. x Conc. | 88 | 27,244.40 | 1.28 |
| Days x Cond. x Conc. | 88 | 15,092.60 | 0.71 |
| Days x Sta. x Cond. x Conc. | 176 | 19,848.00 | 0.93 |
| Error | 528 | 21,330.46 | |

Table 16.--Analysis of variance for 3 x 4 factorial.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 2 | 82,203.50 | 9.00 |
| Concentrations | 3 | 258,168.00 | 25.25 |
| Sta. x Conc. | 6 | 28,693.17 | 3.14 |
| Error | 48 | 9,138.98 | |

Table 17.--Analysis of variance for 2 x 10 factorial.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 1 | 472,060.00 | 15.74 |
| Concentrations | 9 | 31,746.70 | 1.06 |
| Sta. x Conc. | 9 | 54,504.40 | 1.82 |
| Error | 36 | 29,986.90 | |

Table 18.--Analysis of variance for colored baits on white plates.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Baits | 9 | 9,395.00 | 0.58 |
| Error | 54 | 16,097.35 | |

Table 19.--Analysis of variance for colored baits on black plates

| | | | |
|---|---|---|---|
| Baits | 9 | 19,786.11 | 1.81 |
| Error | 54 | 10,908.13 | |

Table 20.--Analysis of variance for black and white contrast study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Backgrounds | 1 | 15,534.00 | 31.46 |
| Error (main plot) | 6 | 374.67 | |
| Plates | 1 | 2,932.00 | 1.18 |
| Backgrounds x Plates | 1 | 704.00 | 0.28 |
| Baits | 1 | 26,139.00 | 10.52 |
| Backgrounds x Baits | 1 | 1,643.00 | 0.66 |
| Plates x Baits | 1 | 4,437.00 | 1.79 |
| Backgrounds x Plates x Baits | 1 | 2,443.00 | 0.98 |
| Error (sub-plot) | 6 | 2,485.00 | |

Table 21.--Analysis of variance for bait shelf life study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Conditioners | 2 | 2,018.50 | 2.31 |
| Days x Conditioners | 6 | 3,178.33 | 3.64 |
| Storages | 1 | 2,898.00 | 3.32 |
| Days x Storages | 3 | 369.67 | 0.42 |
| Conditioners x Storages | 2 | 1,543.00 | 1.77 |
| Days x Cond. x Storages | 6 | 867.00 | 0.99 |
| Error | 30 | 872.83 | |

Table 22.--Analysis of variance for experiment 1 of bait exposure study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 1 | 12,327.00 | 1.70 |
| Concentrations | 4 | 5,560.50 | 0.77 |
| Sta. x Conc. | 4 | 12,788.00 | 1.76 |
| Exposure | 1 | 161,409.00 | 22.23 |
| Sta. x Exposure | 1 | 493.00 | 0.07 |
| Conc. x Exposure | 4 | 17,052.75 | 2.35 |
| Sta. x Conc. X Exposure | 4 | 5,851.50 | 0.81 |
| Error | 8 | 7,261.75 | |

Table 23.--Analysis of variance for experiment 2 of bait exposure study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 1 | 30,016.00 | 2.64 |
| Concentrations | 4 | 52,561.75 | 4.63 |
| Sta. x Conc. | 4 | 35,055.25 | 3.09 |
| Exposures | 1 | 82,140.00 | 7.23 |
| Sta. x Exposures | 1 | 7.00 | 0.00 |
| Conc. x Exposures | 4 | 25,713.00 | 2.26 |
| Sta. x Conc. x Exposures | 4 | 3,621.25 | 0.32 |
| Error | 8 | 11,354.00 | |

Table 24.--Analysis of variance for experiment 3 of bait exposure study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 1 | 595.00 | 0.06 |
| Concentrations | 4 | 3,705.00 | 0.39 |
| Sta. x Conc. | 4 | 4,398.75 | 0.47 |
| Exposures | 1 | 34.00 | 0.00 |
| Sta. x Exposures | 1 | 27,094.00 | 2.88 |
| Conc. x Exposures | 4 | 2,222.75 | 0.24 |
| Sta. x Conc. x Exposures | 4 | 16,895.50 | 1.79 |
| Error | 8 | 9,419.00 | |

Table 25.--Analysis of variance for experiment 4 of bait exposure study.

| Source | d.f. | M.S. | F |
|---|---|---|---|
| Stabilizers | 1 | 1,972.00 | 0.48 |
| Concentrations | 4 | 22,176.25 | 5.38 |
| Sta. x Conc. | 4 | 15,217.50 | 3.69 |
| Exposures | 1 | 59,032.00 | 4.31 |
| Sta. x Exposures | 1 | 28,515.00 | 6.91 |
| Conc. x Exposures | 4 | 23,522.50 | 5.70 |
| Sta. x Conc. x Exposures | 4 | 7,331.25 | 1.78 |
| Error | 8 | 4,125.75 | |

APPENDIX C

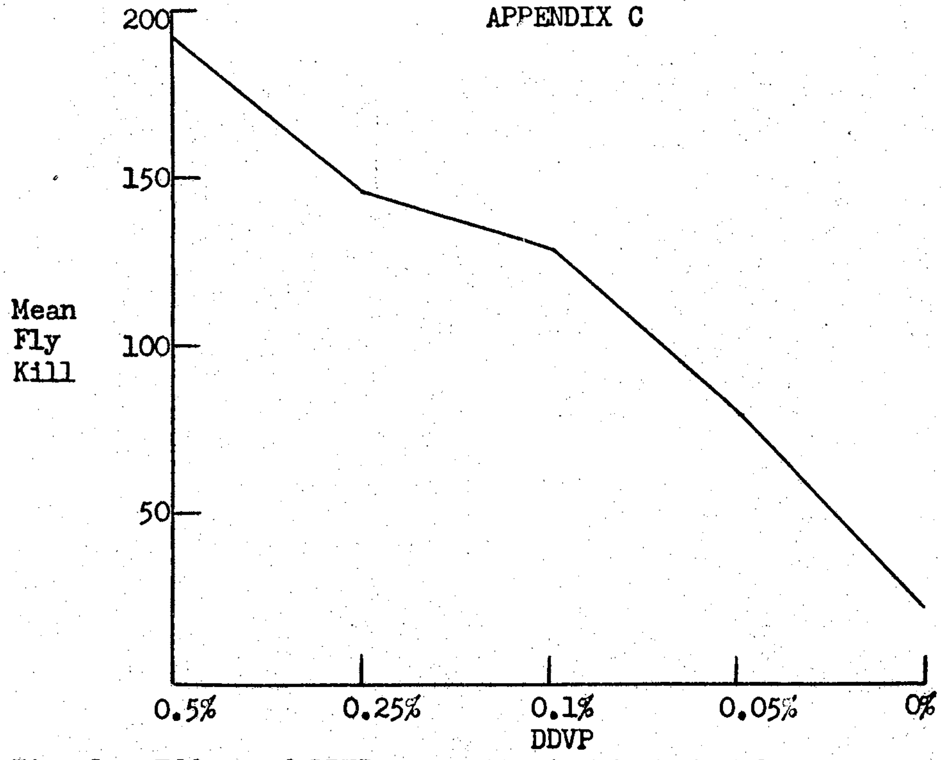


Fig. 1.—Effect of DDVP concentration in baits from table 1 (0.5%, 8-16-10; 0.25%, 5-12-14; 0.10%, 4-11-6; 0.05%, 9-7-13; 0.0%, 20) of preliminary experiment with DDVP and Dibrom, 1961.

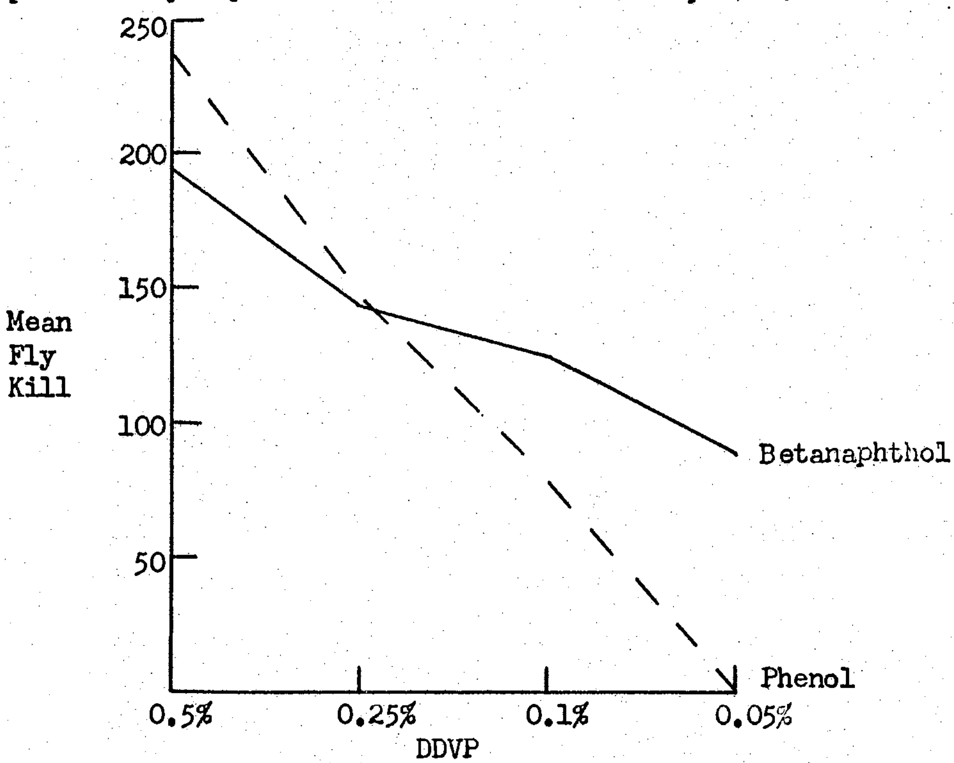


Fig. 2.—Stabilizers x concentrations in baits from table 1 (0.5%, 8-16; 0.25%, 5-12; 0.1%, 4-11; 0.05%, 9-7) of preliminary experiment with DDVP and Dibrom, 1961.

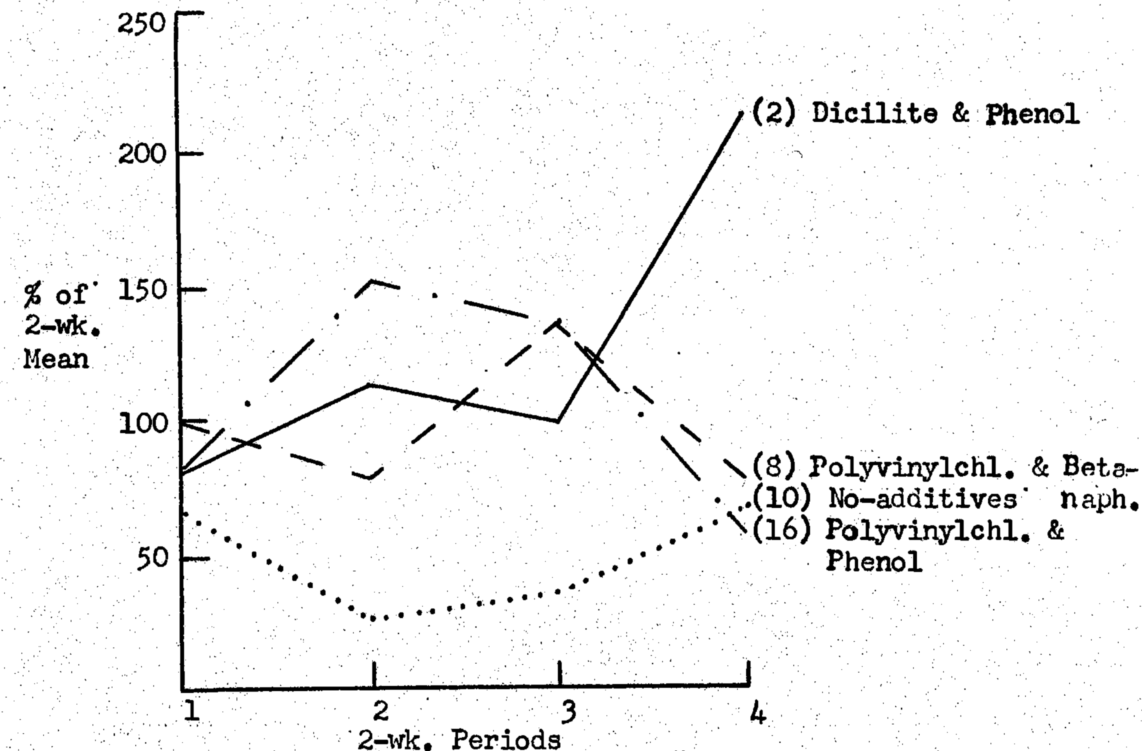


Fig. 3.—2-week periods x 0.5% DDVP baits* of preliminary experiment with DDVP and Dibrom, 1961.

*Formulations given in table 1.

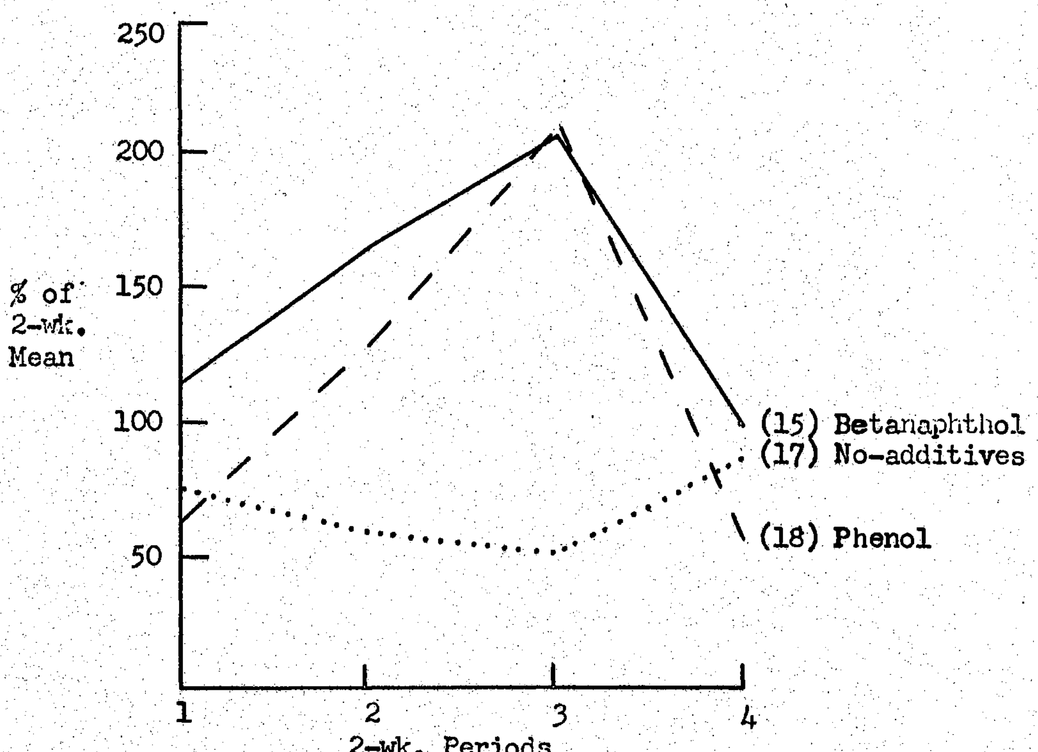


Fig. 4.—2-week periods x 0.5% Dibrom baits* of preliminary experiment with DDVP and Dibrom, 1961.

*Formulations given in table 1.

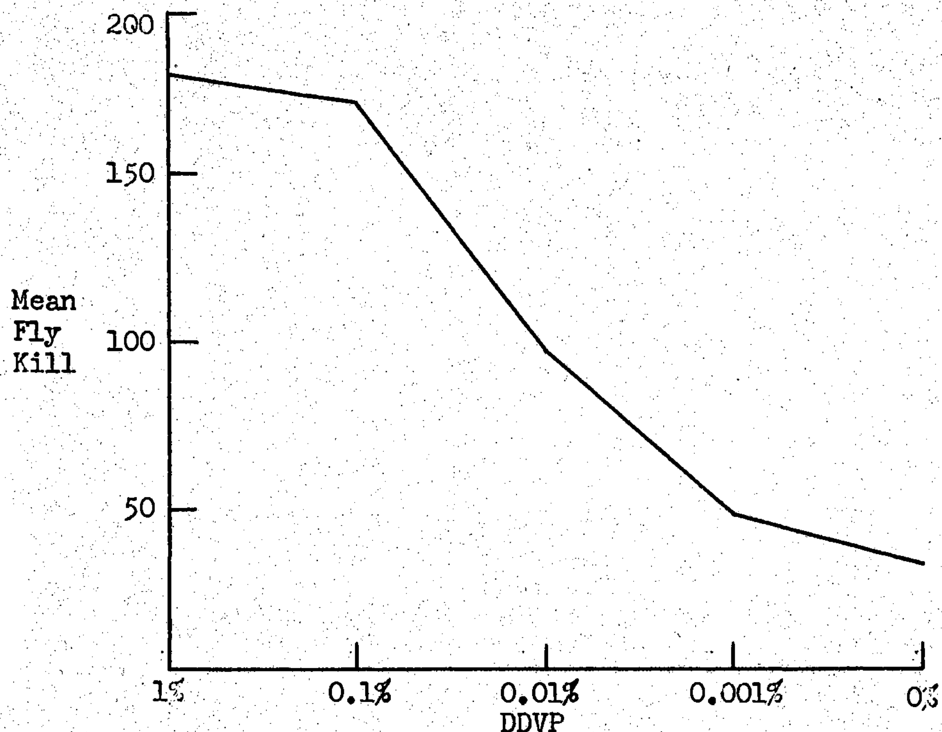


Fig. 5.--Effect of DDVP concentration in 3 x 3 x 5 factorial.

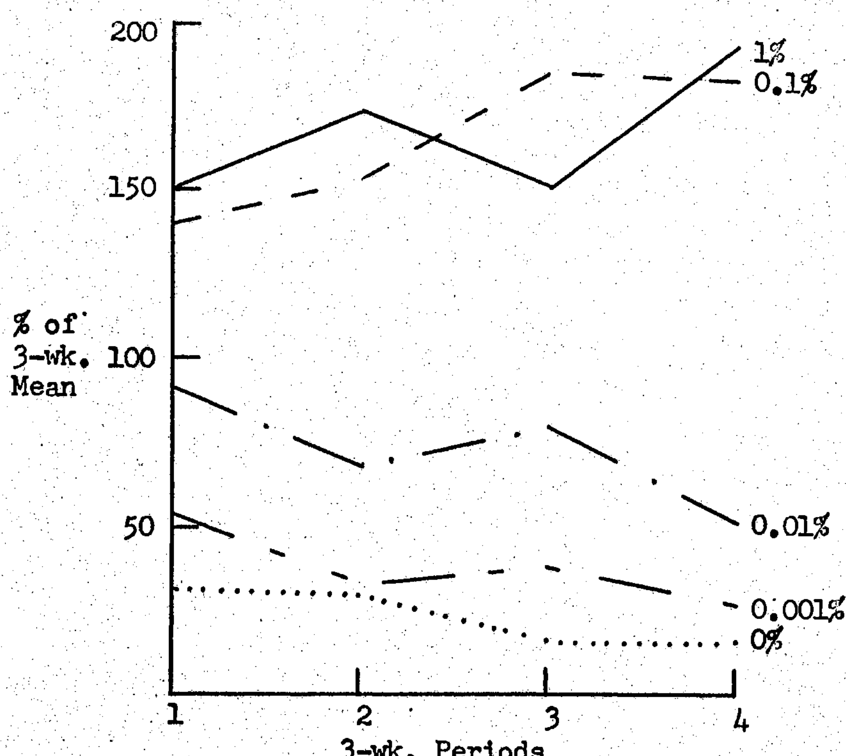


Fig. 6.--3-week periods x DDVP concentrations in 3 x 3 x 5 factorial.

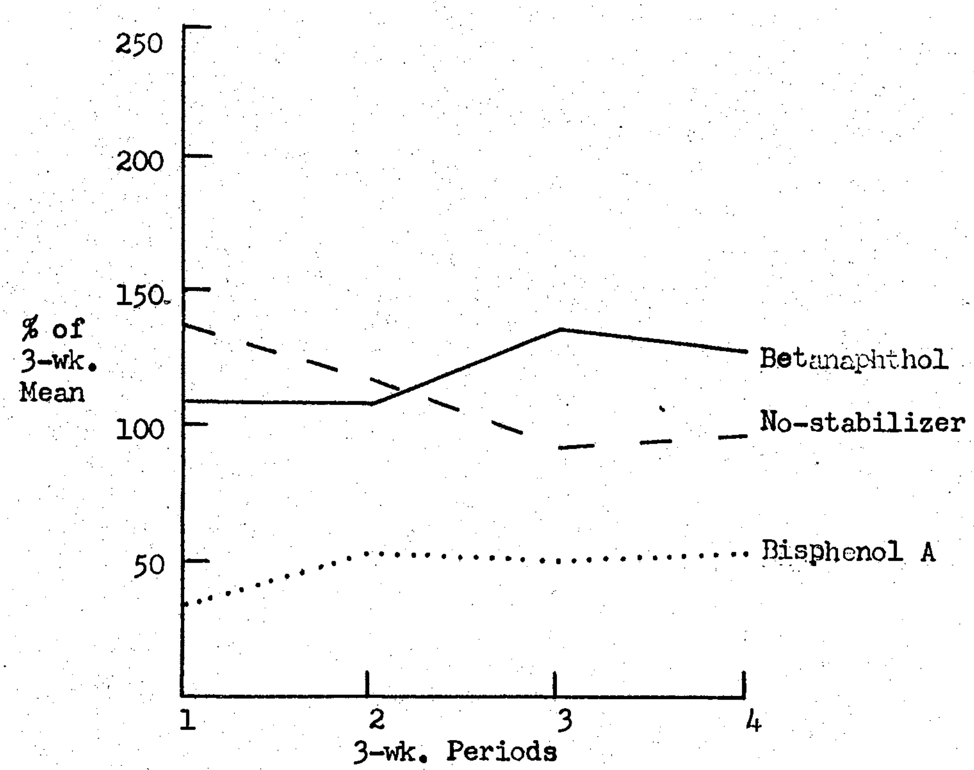


Fig. 7.—3-week periods x stabilizers in 3 x 3 x 5 factorial.

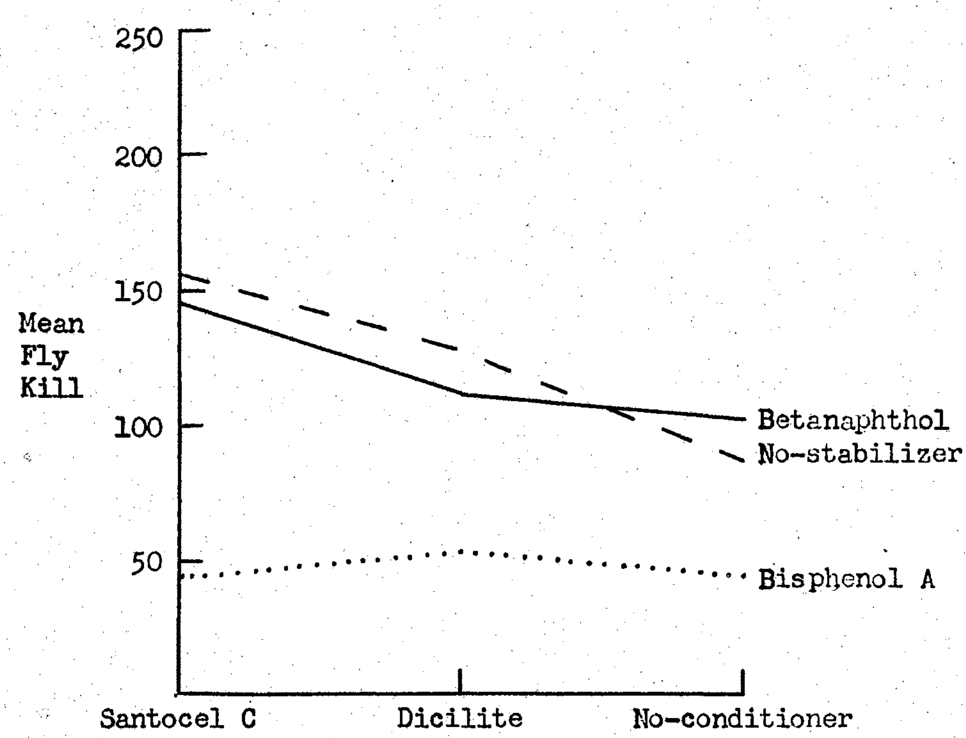


Fig. 8.—Stabilizers x conditioners in 3 x 3 x 5 factorial.

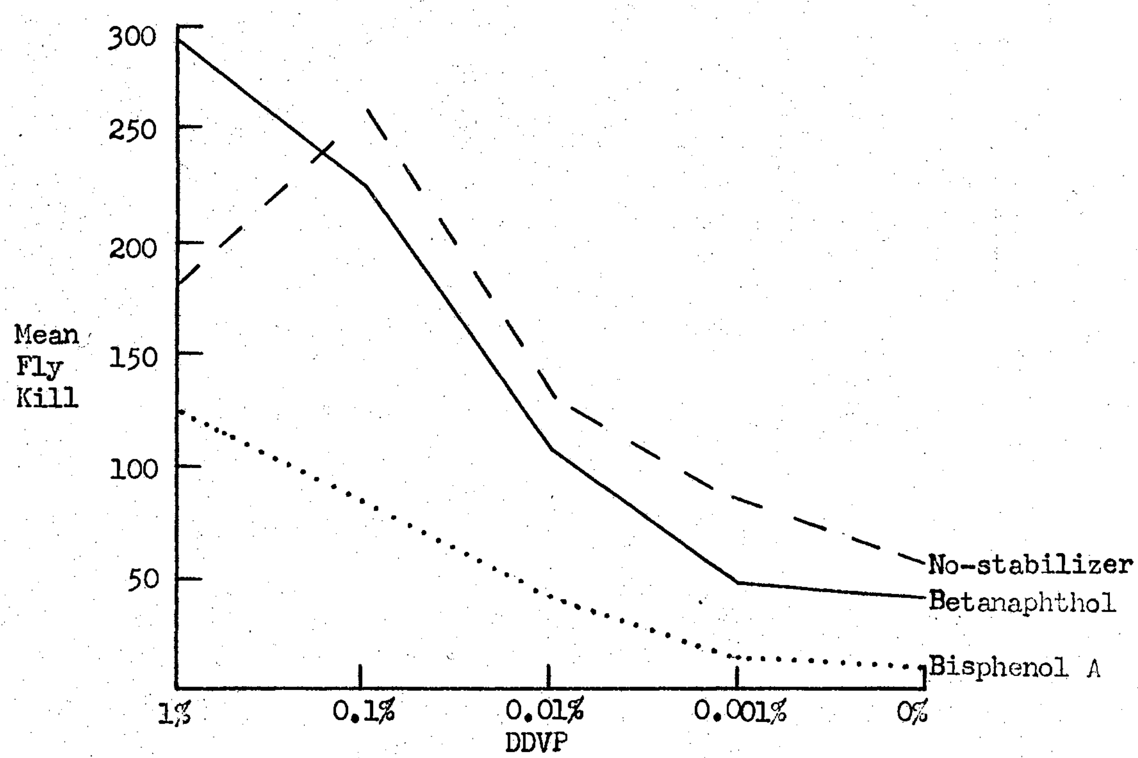


Fig. 9.--Stabilizers x concentrations in 3 x 3 x 5 factorial.

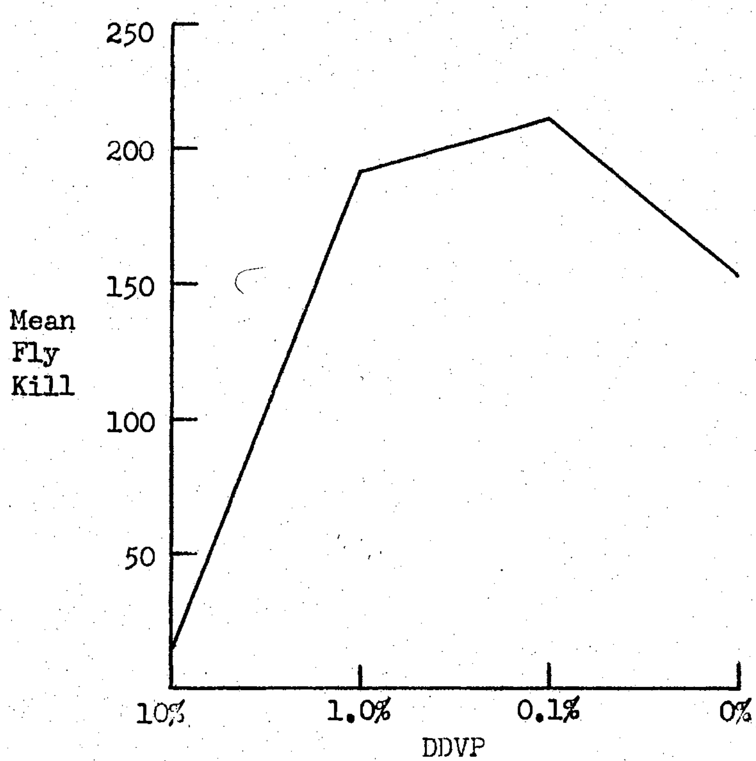


Fig. 10.--Effect of DDVP concentration in 3 x 4 factorial.

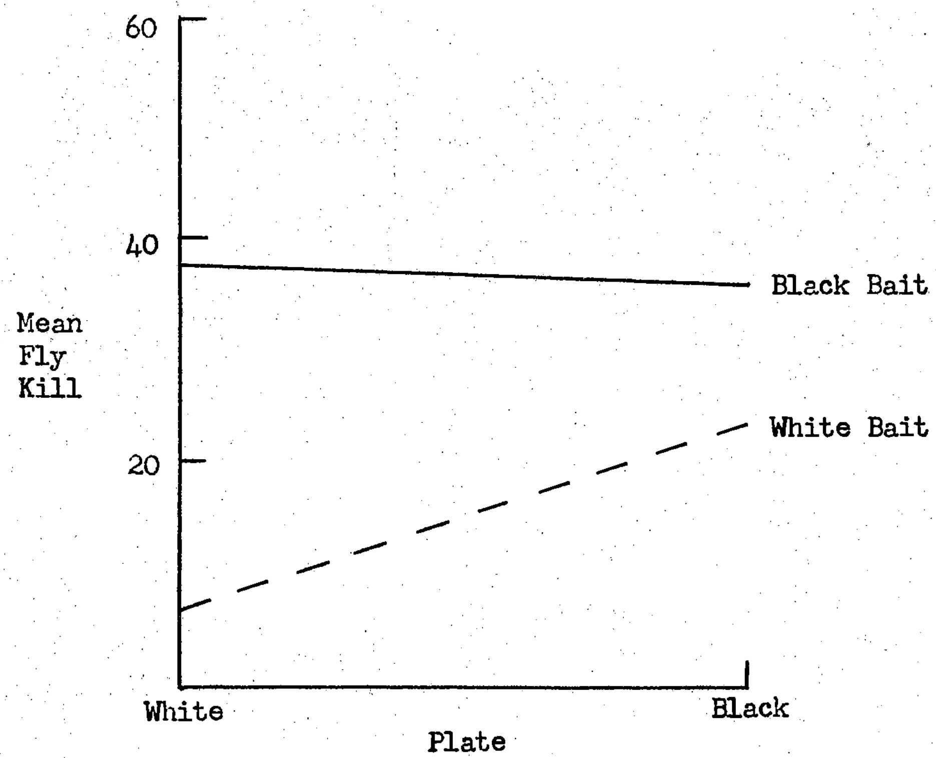


Fig. 11.—Plates x baits in black and white contrast study.

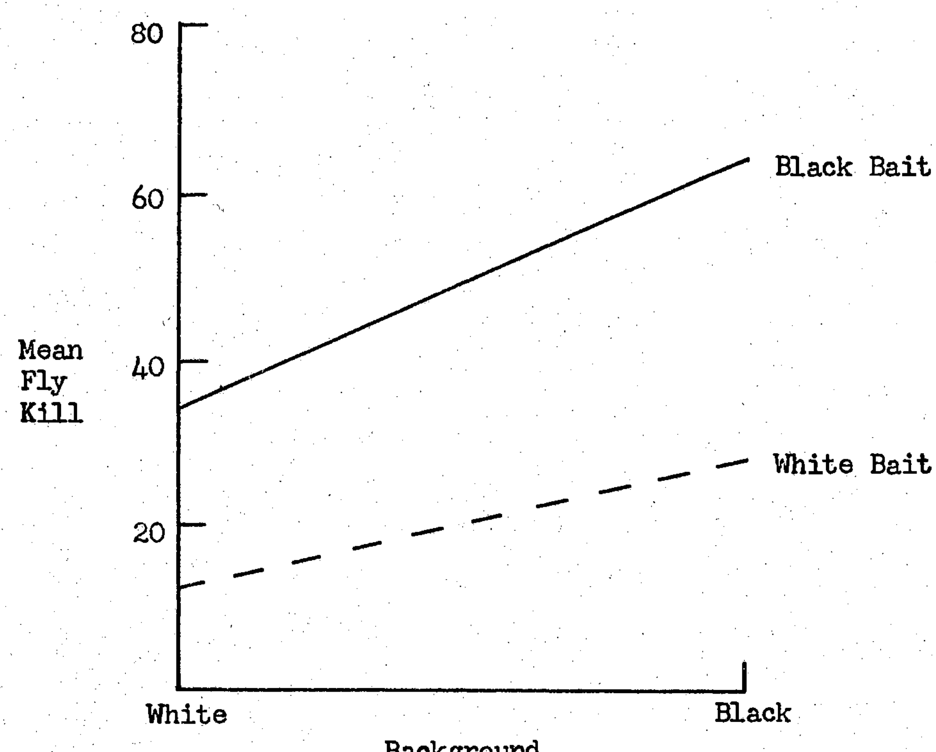


Fig. 12.—Backgrounds x baits in black and white contrast study.

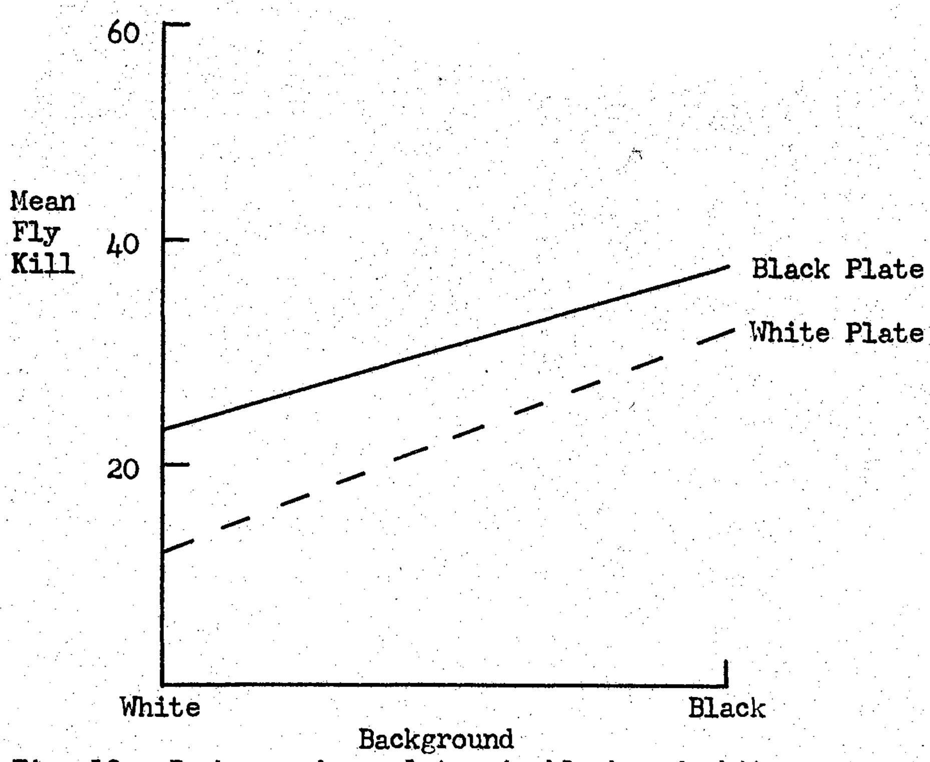


Fig. 13.—Backgrounds x plates in black and white contrast study.

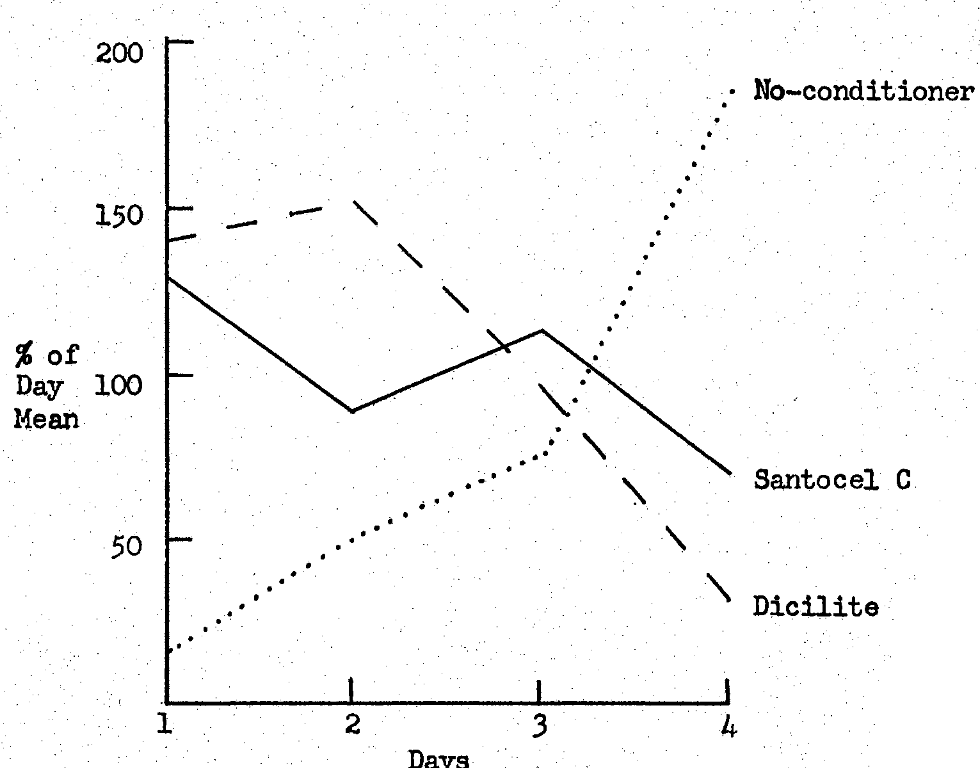


Fig. 14.—Days x conditioners in shelf life study.

VITA

Gary Arthur Mount

Candidate for the Degree of

Doctor of Philosophy

Thesis: THE USE OF FACTORIAL EXPERIMENTS TO DETERMINE THE SENSORY PERCEPTION OF VARIOUS BAIT FACTORS BY Musca domestica L.

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