BIOLOGY AND CONTROL OF THE LESSER MEALWORM: *ALPHITOBIUS DIAPERINUS* A STRUCTURAL PEST IN POULTRY HOUSES

by

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I. INTRODUCTION

The lesser mealworm, Alphitobius diaperinus (Panzer) (Coleoptera: Tenebrionidae), was first cited as a minor pest of stored products. Although this beetle is still considered a stored products pest, the emphasis of its pest status has shifted in recent years. The lesser mealworm has become a problem in certain poultry operations primarily due to the advent of new poultry management techniques. The lesser mealworm probably exists in every major broiler and deep-pit layer operation in the state of Virginia. It has become a structural pest in those houses in which panels of polystyrene or polyurethane are used for insulation. The lesser mealworm bores into and tunnels through these panels forcing the operator to replace them. At the present time, there are no control recommendations for this insect in Virginia.

A clear understanding of which lifestages intiate tunneling behavior as well as the phenological changes that occur in the insulation once tunneling begins, is necessary to postulate reasons why the lesser mealworm bores into insulation. This information will also aid in determining which lifestage to direct control efforts. Baseline

toxicity data (e.g. LD-50's, ect.), using three of the most commonly used insecticides presently approved for use in Virginia poultry houses, were also necessary to obtain. In addition, the residual activity of these materials on polystyrene and plywood surfaces was tested in an effort to determine effective control procedures against lesser mealworm damage.

Although A. diaperinus has become a structural pest in certain deep-pit layer operations, house fly nuisance is the major insect problem in these houses. Because of the extremely large populations of lesser mealworms encountered in the manure of some deep-pit houses, plus the scarcity of information on the subject, predation on house flies by the lesser mealworm is a topic that warranted research.

The objectives of the research presented here are; 1) to follow the chronological events that occur within insulation during the course of a lesser mealworm infestation, 2) to observe the feeding behavior, including food preference, cannibalism, and fly predation of this insect, and 3) to conduct toxicity tests using three insecticides (tetrachlorvinphos, permethrin, and carbaryl) approved for use in Virginia poultry houses.

II. LITERATURE REVIEW

TAXONOMY

Alphitobius diaperinus (Panzer) (Coleoptera: Tenebrionidae) is a small, shiny black beetle belonging to the tribe Ulomini. It has several common names; the lesser mealworm, the darkling beetle, getreideschimmelkafer (corn mold beetle), and the feed bug. Lesser mealworm is the approved common name designated by the Entomological Society of America.

According to Spilman (1966), the generic name Alphitobius has always been associated with the cosmopolitan stored products pests A. diaperinus, the lesser mealworm, and A. laevigatus, the black fungus beetle. The first valid publication of Alphitobius was a listing by Stephens in 1829. A morphological description of the genus was given in Stephens' 1832 Illustrations, giving the type species of the genus as A. picipes. From the original descriptions, illustrations and holotypes examined, Spilman (1966) concluded that A. picipes is most likely a synonym of Alphitobius laevigatus (Fabricius), originally described by Fabricius in 1781 as Opatrum laevigatum. Alphitobius is essentially an African genus. Gebien (1940) listed fifteen

species with their respective synonyms as belonging to this genus. The latest comprehensive work on the genus appeared in an article by Koch (1953). He listed only eleven species, five of which were new species. Koch evidently did not have all the African species listed by Gebien, so his key was not complete (Spilman, pers. letter). The only species of Alphitobius that are considered stored products pests, hence occurring regularly outside of Africa via commerce, are A. diaperinus, A. laevigatus, and more recently, A. viator. Green (1980) published a description and key to distinguish between the adults of these three species. Preiss and Davidson (1970) described the characters for separating late-stage larvae, pupae, and adults of A. diaperinus from those of A. laevigatus.

Due to the many synonymies in the older literature, the taxonomic position of A. diaperinus (Panzer) has been the source of some confusion. Originally described by Panzer in 1797, A. diaperinus has since been given no less than five synonymous specific names (Gebien 1940). This is undoubtedly because its distribution is worldwide. The two most common synonyms in use by some European authors in the 1940's, are A. piceus (Olivier 1792) and A. ovatus (Herbst 1799). Most early authors viewed A. piceus and A. ovatus as separate species, but Bedel, (1906) synonomized both A. piceus and A. ovatus as being the same as A. diaperinus.

Gebien (1920) disagreed, stating that A. piceus Olivier was actually A. laevigatus Fab. and that A. ovatus Hbst. was A. diaperinus Panz. Today it is generally conceded by most tenebrionid taxonomists (Spilman, pers. comm.), that A. piceus (Olivier) is synonymous with the black fungus beetle, A. laevigatus (Fab.) and that A. ovatus (Hbst.) has usually been used to mean the lesser mealworm, A. diaperinus (Panz.). The larval description of A. diaperinus was first given by Schiodte (1878).

NATURAL HABITATS AND WORLD DISTRIBUTION

A. diaperinus has been generally associated with stored products or poultry production, however there have been numerous reports in the literature regarding natural habitats and associations with wild birds and mammals. Most of these reports come from various regions in east Africa. Koch (1953), in his key to the African species of Alphitobius, cited many localitiies in South and Southeast Africa for A. diaperinus. Britton (1940) found Alphitobius larvae within the enclosed confines of an active silver-cheeked hornbill's nest (Bycanistes cristatus) in Northeast Tanzania. A. diaperinus has been recovered from the nests of a kite and a hammerkop in Yiriol, Southern Sudan (Buck 1940) and also on a dead bird in the Red Sea Coast region,

Sudan (Johnson 1933). Guirgis (1971) found A. diaperinus in the nests of cattle egrets (Bubulcus ibis) in the Nile Delta of Egypt.

In addition to birds, A. diaperinus is known to be associated with colonies of bats. Scott (1928) found A. diaperinus within the hollow trunk of a palm tree in occurrence with a dead free-tailed bat (Tadarida spp.) in Portugese East Africa (Mozambique). A. diaperinus has also been reported feeding on bat guano in Ithundu Caves, Kenya (McFarlane 1971), Wad Medani, Sudan (Lewis 1958) and Texas (Davis, et al. 1962).

Other occurrences concerning wild populations of A. diaperinus include its being found in the nests of sparrows (Thompson 1966), pigeons (Levi 1951), starlings (Kemper 1938, Roubal 1917), in rotten stumps (Espanol 1956), under the bark of a dead oak tree (Linsley 1944) and in palm trees and palm fruits (Lepesme 1947). The larvae and adults of A. diaperinus have also been found boring into and living in the scrotum of a moribund Norway rat in the Phillippines (Crook et al. 1981).

A. diaperinus is worldwide in distribution (see Appendix I). If, as some authorities claim, East Africa is its place of origin (Spilman, pers. comm.), then it is not hard to imagine the dissemination of this insect in

foodstuffs aboard trading {hips. It is well established through archeological evidence that from the mid-thirteenth to the fifteenth century the East African coast south to present day Mozambique, harbored flourishing Islamic communities. At that time, these and other Arab seafarers controlled the Indian Ocean trade, exporting mostly ivory, slaves and, undoubtedly, A. diaperinus. Later the Portugese opened East Africa to European commerce (Oliver and Fage 1962). Indeed large populations of A. diaperinus have been reported living in the garbage dumps near docks {O'Mahony 1950). A. diaperinus was found in Japan as early as 1875 (Marseul 1875), over 20 years after Admiral Perry and his Black Fleet opened Japan to world trade. In 1891, Fowler reported A. diaperinus thriving in the "deep hot coal mines of the Northumberland district (England), where it abounds in the stables, having probably been introduced in some of the horse's food." It is interesting to note that the first references regarding A. diaperinus as a stored products pest were from Senegal in stored ground nuts (Rouband 1916) and from Barbados in decaying cotton seed (Bovell 1917); both geographic points along the transatlantic slave route.

STORED PRODUCTS

A. diaperinus, as a pest of stored products has been associated with products of high moisture content (copra, oilseeds,etc.) or with grain or grain by-products that have become moldy or slightly out of condition. From an investigation using eight types of grain, Sarin and Saxena (1975) found that larvae preferred wheat, while adults preferred cowpea and that the majority of the grains tested were attacked at the germ end. For a complete list of commodities which A. diaperinus is known to infest, see Appendix II. A. diaperinus has also been reported occurring in and around mausoleums, crypts and cemeteries (Ebeling 1975).

ASSOCIATION WITH POULTRY

Although it has been reported occurring in the manure of cows (Toyama and Ikeda 1976, Legner and Olton 1970), horses (Legner and Olton 1970), and pigs (Tor'ch 1979, Legner and Olton 1970), A. diaperinus, as a livestock pest, is most commonly found in the manure of poultry. A. diaperinus was not reported occurring with poultry production in large numbers until the early 1950's. It was at this time that poultry production began its rapid growth

as an agricultural industry in the United States. At first the different phases of the industry used essentially the same management techniques which consisted primarily of penned areas on a dirt floor. Gradually, different management techniques evolved for various systems of poultry production such as meat production, egg production, and breeder flocks.

Breeder flocks today are generally housed in structures where a portion of the floor space is raised and slatted, thereby separating the birds' roosting area from the droppings thus allowing the manure to dry. A. diaperinus has never been recorded occurring in this type of operation, but it has been observed in some breeder houses (Weaver and Ruszler 1981).

Present commerical egg production use wire cages which effectively separate the birds from their droppings, allowing the manure to accumulate underneath. The common practice is to suspend the cages ca. 1 meter above the floor. The newer high-rise or deep pit houses are two-storied structures in which the upper story is used for housing the birds and the lower story for the accumulation of manure. Pfieffer (1954) and Pfieffer and Axtell (1980) found A. diaperinus to be one of the most abundant coleopteran species in both types of houses. Because

cleanout is much less frequent, enormous populations of A. diaperinus can develop upon the manure cones of high-rise houses. Recently A. diaperinus has caused concern as a structural pest because it bores into and tunnels through exposed insulation within these houses (Spilman 1968, Gall 1980, Smith 1981). In addition, when the manure is cleaned out of a high-rise operation and piled or spread about the fields, the adult beetles have been known to take flight en masse and invade dwellings and yards up to five or ten miles away (Thornberry 1978, Jerrard 1980, Smith 1981).

Birds are kept upon a litter-covered floor in meat producing operations (broilers, brooders, and turkeys). The concept of a built-up litter, whereby fresh dry litter is merely added over old moist litter, became widespread practice (McCreary and Catts 1954). The practice of built-up litter also allowed for a buildup of lesser mealworm populations. Lancaster (1969) found A. diaperinus to be the most common insect in poultry litter. The present standard sidewall construction for broiler/turkey houses in this country involves fiberglass insulation covered with a sheet of polyethylene plastic (vapor barrier) behind 3/4" plywood (Weaver and Ruszler 1981). A. diaperinus causes little structural damage in these houses. However in Japan (Ichinose, et al. 1980), France (Chaix 1980), and New

Zealand (Dale, et al. 1976), where polyurethane and/or polystyrene is used, structural damage has been reported in broiler houses. A. diaperinus were first reported in large numbers in U.S. poultry operations by Gould and Moses (1951). Harding and Bissel (1958) reported the larvae of A. diaperinus in a brooder house feeding upon the flesh and internal organs of dead and dying chicks. The voracity of the lesser mealworm was affirmed by Harris (1966) who reported several hundred adult beetles attacking and devouring a ten inch rat snake. He also reported A. diaperinus attacking the eggs and pupae of Tenebrio molitor L., reducing the colony to but a few individuals.

VECTOR POTENTIAL

A. diaperinus is an intermediate host for the pinworm Subulura brumpti Lopez-Neyra, a common cecal nematode of chickens and turkeys (Alicata 1939). The pathological symptoms in poultry are mild (Cuckler and Alicata 1944). A. diaperinus has also been reported as an intermediate host of the funnel-shaped tapeworm, Choanotaenia infundibulum (Case and Ackert 1940, Elowni and Elbihari 1979), and the broad-headed tapeworm, Raillientina cesticillus (Case and Ackert 1940). Both of these tapeworms infest the jejunal region of

the small intestine in poultry (Hofstad et al. 1972) and cause similiar symptoms. A. diaperinus was found to be an unsuitable intermediate host for the tapeworm Hymenolepis nana Von Siebold (Preiss 1969).

Eugenio, et al. (1970) demonstrated the ingestion and retention of the mycotoxin F-2 in A. diaperinus throughout metamorphosis, starvation, and after death. This indicates that the insect may be a potential carrier of the fungal toxin to food and feeds. Natural populations of A. diaperinus in the poultry house show a significant harborage of Aspergillus flavus Link, the main fungal species responsible for aflatoxicosis in poultry (De las Casas, et al. 1972, Harein and De las Casas 1969). In laboratory life history studies, Wilson and Minor (1969) observed a direct correlation between Aspergillus fungal growth on semi-synthetic media and optimal larval development.

Surface-disinfected A. diaperinus were shown to be capable of harboring Escherichia coli Migula, causative agent for avian colibacillosis, and Salmonella typhimurium Loeffler, the most frequent Salmonella species implicated in parathyroid infections (De las Casas, et al. 1968). Harein et al. (1970) isolated five species of Salmonella and forty-eight serotypes of E. coli, twenty-six of which are known to be pathogenic for animals or man, from natural

populations of lesser mealworms in a brooder house. However, when De las Casas, et al. (1974), using allantoic inoculations of chicken eggs, tested E. coli serotypes that had been recovered from within mealworms, they found a reduction in virulence when compared to similiar serotypes recovered from poultry. When comparing surface litter, subsurface litter, dust, and surface-disinfected A. diaperinus collected from a turkey brooder house, Harein, et al. (1972) found that although the lesser mealworm carried the highest percentage of E. coli, it carried the least total number of bacteria per area. These results failed to prove A. diaperinus as a significant source of microorganisms in the poultry environment compared to the other substrates tested.

A. diaperinus has been implicated in a number of viral diseases of poultry. Eidson, et al. (1965) conducted preliminary experiments and later (Eidson, et al. 1966) proved that A. diaperinus was capable of transmitting Marek's disease, a DNA herpes virus that for years was the poultry industry's most costly disease (Schmidttle 1966). Interest in A. diaperinus skyrocketed during the late 1960's but has subsequently receded with the advent of a vaccine for Marek's disease in 1972. The causative agent of Gumboro disease, a highly contagious viral disease affecting young

birds, has been isolated from A. diaperinus collected 8 weeks following a serious outbreak in Delmarva broilers (Snedeker et al. 1967). The virus causing infectious bronchitis, Massachusetts strain, could not be re-isolated from orally-infected A. diaperinus after four hours post-infection (Stuke and Kaleta 1970). A. diaperinus was also shown to be an ineffective carrier for Reovirus 24 (De las Casas, et al. 1973), fowl pox and Newcastle's disease (De las Casas et al. 1976).

BASIC BIOLOGY

Workers developed their own synthetic and semi-synthetic rearing media for rearing A. diaperinus. This may explain some of the differences in the results they reported. Lancaster and Simco (1967) used four different diets at temperatures ranging from 15.5°C to 21.1°C and found average egg-to-adult lifespans of 39.2 days, 47.3 days, 55.6 days, and 79.3 days. Barke and Davis (1969) found the lifespan to be between 60-85 days at 21°C. Wilson and Minor (1969) found the optimal development temperature to be 32.2°C (90°F) resultng in an average egg-to-adult lifespan of 45.6 days. At higher temperatures (37.8°C), developmental time was shorter (42.3 days) but mortality was

used usually higher. There was no development at temperatures below 10°C.

Wilson and Minor (1969) and Preiss (1969) presented physiological descriptions of the egg and reported egg dimensions ranging from 1.2-1.3mm in length and 0.5-0.7mm in width. Lancaster and Simco (1966) reported the incubation time to be 4.3 days at 18.3°C, but as Preiss pointed out, these results were probably erroneous since the oviposition sites were fermenting wheat kernels and, because of heat given off in the process of fermentation, the microclimate of the incubating eggs was undoubtedly much warmer than 18.3°C (65°F). Barke and Davis (1969), using relative humidities of 70, 80, and 90%, found little difference in incubation time and only a slight increase in hatchability at 70% RH. Preiss and Davidson (1969) found that temperature, not relative humidity, is the crucial environmental factor governing both incubation time and hatchability of A. diaperinus eggs, optimal temperature being 26.7°C (80°F). At constant temperatures above 37.8°C (100°F) or below 10°C (50°F), there was no hatch at 25% RH. Ichinose, et al. (1980) showed that exposure at 7°C for five days will reduce hatchability to 2%.

Lancaster and Simco (1966) reported the larval stage of A. diaperinus lasting an average of 48.5 days and having

10-12 instars. Barke and Davis (1969) reported 6-9 larval instars and gave the head capsule measurements for instars 1-6. Wilson and Minor (1969) published an excellent description of the various larval instars of A. diaperinus with regard to size, coloration, and developmental duration. They reported up to 11 instars, depending upon temperature, with instars 7-11 being the result of of lower temperatures, having slower developmental times, and displaying little difference in size. Preiss (1969) defined the activity threshold temperature for larval, as well as adult A. diaperinus, as 7.2°C (45°F). Below this temperature they became incapacitated. A characteristic tunneling behavior throughout the agar-based medium was observed to occur in all larval instars except the first. A distinct prepupal stage, lasting 3-10 days was described by Wilson and Minor 1969. Using the criterion of percent weight gain, Preiss (1969) found no significant difference between individually-reared and group-reared larvae. However he did find a significant increase in percent mortality in the group-reared larvae. In the absence of food and water for over 10 days, middle instar larvae suffered a 97.3% mortality rate, whereas only 5% of the late instars tested died; the other 95% metamorphosed to the pupal stage (Preiss 1969). Most late-instar larvae preferred burrowing into soil (Preiss

1969) before assuming the slightly curled position of the inactive prepupal stage.

Lancaster and Simco (1966) reported the pupal stage lasting an average of 6.9 days, while Barke and Davis (1969) reported an average duration of 9.2 days. Preiss (1969) reported that pupal duration was related to temperature, with the shortest duration being 3.9 days at 37.8°C (100°F) and the longest being 12.1 days at 21.1°C (70°F). Chilling the pupae at 0.5°C (33°F) for 2-4 days increased the incubation period. Five days or longer at this temperature resulted in non-emergence (Preiss 1969).

The teneral adult has been described as soft-bodied and reddish brown in color, but hardens and becomes a shiny black in about three days (Wilson and Minor 1969, Barke and Davis 1969). Sex ratios in the laboratory varied; Barke and Davis (1969) reported 1:1, whereas Wilson and Minor (1969) reported 1:2.25. Preiss (1969) reported the sex ratio to be erratic in laboratory colonies but in the field he found a preponderance of males to females. The preoviposition period for A. diaperinus reported in the literature varies, ranging from an average of 10.8 days (Lancaster and Simco 1966) to an average of 12.7 days (Preiss and Davidson 1969). Wilson and Minor (1969) reported that the preoviposition period bears little relationship to temperature. Eggs are

laid singly or in clusters. Preiss and Davidson (1969) reported that healthy females are apparently able to store sperm and deposit viable eggs throughout their life, with an average egg production of 3.5 eggs/day. Wilson and Minor (1969) found egg production to be temperature dependant, with highest production (5.5 eggs/day) occurring at 32.2°C (90°F). They speculated that adult A. diaperinus probably live more than a year. Preiss and Davidson (1969) found the average adult lifespan to exceed 400 days, with one pair of beetles living in excess of 700 days. Preiss (1969) showed that adults are much more resistant to starvation than the larvae, and are able to survive an average of 19 days without food or water. He reported no significant difference in resistance to starvation between males vs females or newly-emerged vs older beetles.

Pfeiffer and Axtell (1980) sampled shallow-pit and deep-pit caged layer houses in the three zoogeographic regions of North Carolina. They found that A. diaperinus larvae were usually more numerous than the adults. Both larvae and adults were more abundant in the Piedmont region and least abundant on the Coastal Plain. Population peaks varied according to region: November for the Coastal Plain; July-September for the Piedmont; and September-early October in the Mountains. Larval populations peaked earlier in the deep-pit houses than in the shallow-pit houses.

Significantly higher numbers of adults were present during the winter months, indicating that the adult stage may be the primary, but not the exclusive, overwintering form.

The seasonal distribution of lab-reared A. diaperinus was investigated by Sarin (1978). She found that the adult population peak in late August was not in phase with the larval population peak, which peaked earlier in May. When conditions were unfavorable, older larvae were observed to feed upon younger larvae.

MORPHOLOGY AND BIOCHEMISTRY

Sexual dimorphism is present in A. diaperinus. Hewlett (1958) and later Halstead (1963) described a method of separating the sexes on the basis of the curvature of the mid- and hind-tibial spurs. In the female both spurs are straight, in the male one spur is straight while the other spur is recurved inward at its apex. However, with live insects this method is both laborius and time-consuming. Barke and Davis (1967) described another method involving gentle pressure being applied with a blunt instrument to the ventral surface of the insect's abdomen forcing the genitalia to be extruded. Sexual dimorphism was also found to exist in the pupal stage with the female possessing a

noticeable, fleshy pair of second valvifers. These are absent in the male pupae. It has been noted by several workers that both female pupae and adults are generally larger than the male.

The external morphology of the head region was described by Pajni and Bandlish (1972) and Sarin, et al. (1968). A description of the mandibles based on scanning electronmicroscopy has been published by Kvenberg (1977). Micromorphology of the elytra shows A. diaperinus to have a regular polygonal pattern common to the family Tenebrionidae (Khalaf 1980).

The internal anatomy of A. diaperinus was presented by Vaidya and Tembe (1957). An extensive Malpighian tubule network, together with the presence of rectal pads indicates that adult A. diaperinus are capable of some water conservation. An illustrated description of the spermathecal structure was presented by Surtees (1961). Satija, et al. (1977a) described the morphological changes in the brain of A. diaperinus throughout its life history. Satija, et al. (1977b) also published a similiar treatise regarding visual centers.

Several workers have reported the appearance of posterior scent glands in adult lesser mealworms. Wilson and Minor (1969) noted that the oily fluid produced by these

glands had a "musky odor and caused sexual excitement in both sexes." Tseng, et al. (1971) described the morphology and chemistry of this odoriferous gland and reported no difference in morphology or secretion composition between males and females. The two major constituents of the secretion were found to be benzoquinone compounds.

A. diaperinus has been found to a convenient insect with which to conduct biochemical analysis because of its size, relative ease of maintaining large laboratory colonies, and its status as a stored products pest. Sharma, et al. (1973) have described the karyotype (2n=19) and sex mechanism (XO) of the adult male A. diaperinus. Sarin (1973a) found gut enzymes suited for both phytophagous (maltose, amylase) and carnivorous (pepsin, trypsin) feeding habits in both larval and adult A. diaperinus. Sarin (1973b) found maximum amino acid concentration of hemolymph to occur in the final instar and pupal stage, with a subsequent drop in these concentrations following metamorphosis. Lockey (1979) found that A. diaperinus could be distinguished from three other genera of tenebrionids which infest stored products by comparison of their cuticular hydrocarbons.

CONTROL

Saxena and Sarin (1974) found the LC-50 values and relative toxicities of five insecticides against four day old adult A. diaperinus. Mevinphos was shown to be the most toxic, followed by fenitrothion, pyrethrum, lindane, and p,p' DDT. Blahutiak and Barus (1970) reported the action of five insecticides on the lesser mealworm. Hewlett (1962), using topical application on the closely related species, Alphitobius laevigatus, reported the LD-50 values for n-valone, DDT, dieldrin, and allethrin. He also found in each case with equal dosages, that there was increasing toxicity with decreasing volume of solution.

Silbermann (1967) performed toxicity tests in the laboratory which confirmed results obtained from spray tests in actual broiler operations. He found that ronnel applied at rates of 0.5, 1.0, and 1.5 pounds per 1000 sq. ft.; dimethoate at 1.0; chlorpyrifos at 0.5 and 1.0; and carbaryl at rates 1.0 and 1.5 pounds per 1000 sq. ft. all gave satisfactory control. The carbaryl treatment had the longest residual activity. Methoxychlor proved to be ineffective. With the exception of chlorpyrifos, all the compounds lost toxicity when combine with fuel oil and cresylic acid. Simco, et al. (1966) treated the litter in eight different broiler houses. He found that spraying with

either 0.5% carbaryl, 0.5% ronnel, or 1% malathion after an intial cleanout, then treating the litter after each successive flock of broilers with 5% carbaryl dust at a rate of 2.5 pounds per 100 sq. ft. would greatly reduce the numbers of lesser mealworms. Spraying with 0.5% dichlorvos or 0.35% coumaphos however, proved to be ineffective. To prevent reinfestation of the broiler house from beetles emerging from discarded litter, Bray, et al. (1980) recommended dusting the old litter with 5% carbaryl at a rate of 2.5 pounds per 100 sq. ft. before cleaning out. Prior to the addition of new litter, fumigating with formaldehyde for 24 hours; applying one gallon 50:50 mixture of cresylic acid:fuel oil per 1000 sq. ft.; and dustng new litter with 5% Sevin as before, was suggested as a control program. Swatonek (1970) achieved satisfactory control upon cleanout by spraying with lindane, taking care to treat the cracks between floor and woodwork. Lancaster, et al. (1969) investigated the potential of pre-treated rice hull litter for the control of the darkling beetle. In tests using plastic containers filled with insecticide-treated rice hulls, chlorpyrifos, ronnel, and mobam showed 100% effectiveness for up to eight weeks, whereas coumaphos and malathion were less effective. Carbaryl proved to be the least effective material. Storage deterioration of all

materials tested was negligible. However, in experimental plots containing broilers, there was a considerable reduction in insecticidal activity, presumably due to chemical degradation or dilution because of feed spillage and accumulation of droppings. Only chlorpyrifos was effective for the entire eight week trial period and ronnel lasting three weeks, was by far the second most effective. No insecticide residues were found in the birds' skin, muscle, or liver, and not more than 0.02 ppm was detected in the fat of birds grown for eight weeks on litter pre-treated with 0.125% chlorpyrifos. Gall (1980) recommended dusting the litter with tetrachlorvinphos 50% WP at a rate of 4 pounds per 5000 sq. ft. every six months to prevent lesser mealworm problems in broiler houses.

Alicata (1945) found that a mixture of crude napthalene and sand spread over the surface of manure at a rate of about 5 grams per sq. ft. killed many adult A. diaperinus, but some were able to survive under encrusted droppings. In caged layer operations, Kartman, et al. (1950) noted that A. diaperinus and other burrowing arthropods create a second layer beneath the surface manure, composed of manure and earth (subfaex) in which all lifestages of A. diaperinus may be found. Under simulated natural conditions they found benzene hexachloride and parathion, when applied to the

manure, to be more effective than methoxychlor, chlordane, toxaphene, or DDT. They noted that control "apparently resolves itself into a question of spreading and penetrating ability rather than one of comparative toxicity". In empty layer houses, chlorpyrifos 2%, and in inhabitated houses, Permenant (permethrin and pyrethrum) 0.4% have successfully controlled the getreideschimmelkafer, A. diaperinus, in Germany (Heimbucher and Kutzer 1979). Fenvalerate 0.5%, fenchlorphos 2%, and Lysizid S (malathion + novathion + pyrethrum) were less effective. Thornberry (1978) suggested that spraying walls and frameworks with carbaryl when beetle populations are high would prevent damage to insulation.

In Hungary (Nemeseri and Gesztessy 1973) and Germany (Geissler and Kosteis 1972) satisfactory reduction in beetle populations has been achieved without the use of insecticides by opening the houses to winter temperatures for at least five days inbetween flocks. Ichinose, et al. (1980) found that paper and aluminum foil coverings effectively prevented boring by A. diaperinus into polyurethane, polystyrene, and fiberglass insulation.

III. INSULATION INFESTATION STUDIES

Introduction

The lesser mealworm, A. diaperinus, has always been associated with poultry and probably exists in almost every poultry unit where reasonably dry litter or manure is allowed to accumulate. However, it was not until large-scale poultry production in the 1950's, with concurrent changes in management techniques, did the lesser mealworm become a concern to poultry operators. Manure and litter cleanout has become much less frequent, resulting in enormous populations of A. diaperinus developing within poultry houses. The lesser mealworm has become the most abundant beetle inhabitating litter and manure in both the broiler and egg industry (Lancaster, et al. 1969, McCreary and Catts 1954, Pfieffer, et al. 1954, Pfieffer and Axtell 1980).

Polystyrene or polyurethane panels, presently used in the interior of many high-rise layer houses as insulation, are often destroyed at an alarming rate as the result of tunneling activity by the lesser mealworm (Spilman 1968, Gall 1980, Smith 1981). Ichinose, et al. (1980) showed that A. diaperinus would infest polyurethane foam, expanded

polystyrene and spun fiberglass. Only late instar larvae were found to intiate tunneling and all lifestages failed to survive on these materials. A preponderance of adults was found to occur within the tunnels of infested polystyrene insulation in a deep-pit layer operation in Washington Co., Virginia. It remained unclear whether the adults encountered in field infestations of insulation were the result of metamorphosing larvae or whether adults from the media, along with late instar larvae, also were tunneling into the panels. There existed the possibility that a chronological succession of lifestages was occurring within the insulation during the course of these infestations.

Objectives

The objectives of these studies were 1) to follow the events occurring within the insulation over time during a laboratory infestation of polystyrene and 2) to compare concurrent infestations by the lesser mealworm into polystyrene, polyurethane, and fiberglass to determine if preferences for one type of insulation over another type exist.

Materials and Methods

Samples of A. diaperinus were collected from a large field population thriving on the manure cones inside a deep-pit poultry house in Amelia Co., Virginia several months before the test was to be conducted. A. diaperinus, along with the surrounding media and microfauna, were put in five 19-liter plastic buckets. The stock colonies were housed in a cabinet at room temperature (ca. 21°C) and under 24-hour darkness.

Polystyrene and polyurethane panels of uniform size were cut by handsaw and the styrofoam sectioning apparatus shown in Figure 1. The sectioning apparatus consisted of a medium gauge metal guitar string kept under tension by 2 guitar head tuners at both ends of a 2" x 4" board two feet long. A Powerstat variable autotransformer (Superior Electric Co., Bristol, Conn. U.S.A.) was hooked up at the opposite ends of the wire string throughwhich an adjusted current of ca. 8-9 volts was conducted. The current was of sufficient strength to heat the wire and section the styrofoam at the desired thickness. The wire was raised approximately 0.6cm from the backboard.

Study 1. The first study investigated infestation by A. diaperinus into polystyrene or styrofoam insulation. The styrofoam insulation consisted of white closed-cell expanded

polystyrene. Seventy-five panels of uniform size (ca. 15cm x 8cm x 0.6cm) were cut by handsaw and the styrofoam sectioning apparatus. Fifteen of these panels were placed lengthwise in the media of each of the five replicate stock colonies. At intervals of two to three days following the introduction of the panels, one panel from each stock colony was randomly selected and removed. Any beetles or larvae crawling about on the surface of the panels were brushed back into their respective replicate colony and the panels of each replicate were placed immediately into seperate "stop baths" of 90% ethanol. This halted all activity within the insulation. After 10 minutes the panels were removed and allowed to dry. Each alcohol bath was strained through a No. 50 wire mesh screen and the recovered organisms were recorded and counted. Each panel was then sectioned into 0.6cm strips and examined under the stereoscope. The number of eggs, larvae, pupae, and adults of A. diaperinus was recorded, as well as the presence of any other arthropods. A. diaperinus larvae were recorded as either early, middle, or late instars in accordance to head capsule width. The test was conducted for 35 days.

One week upon completion of this test, four fresh polystyrene panels of the same uniform size were introduced into each replicate of the stock colonies. The panels were

removed on 1, 2, 4, and 6 days post-introduction, sectioned, and the recovered lifestages were counted and recorded as previously described. This was done to corroborate early trends observed in the intial infestation of the insulation.

Study 2. A similiar insulation infestation study was conducted to include the three major types of insulation used in Virginia poultry houses. The three types of insulation used were:

1.) Extruded polystyrene insulation formulated in small open cells within a filler matrix and blue in color. It was cut into panels approx. 12cm high x 10cm wide by 1 cm thick;

2.) polyurethane insulation formulated in small open cells, no filler material, and yellow in color. It was cut into panels approx. 12cm high by 10cm wide by 1cm thick;

3.) spun fiberglass pink in color and packed together in a cylindrical fashion by pint ice cream containers without tops or bottoms, 8cm in diameter, and cut to a height of 2.3cm.

Eighteen samples of each insulation type were prepared. Each insulation sample was weighed prior to introduction into the stock colonies to determine weight loss or weight gain due to the tunneling activity of A. diaperinus. The

average weights of the polystyrene, polyurethane, and fiberglass samples were; 4.10gr (+ 0.51), 3.14gr (+ 0.45), and 5.96gr (+ 0.38) respectively. A sham sectioning was performed on three polystyrene and three polyurethane panels and the panels were weighed to determine if a correction factor was needed to compensate for any weight difference that might have been due to the sectioning procedure. There was no measurable difference in weight between the intact (pre-sectioned) and sectioned panels.

Three replicate stock colonies of A. diaperinus, kept in a growth cabinet at a temperature of 28.3° ± 1.6°C and 24-hour darkness, were used in the test. Six of each of the three types of insulation being tested were placed lengthwise into the media of each colony. One piece of gach type of insulation was randomly selected and removed from each replicate stock colony at 1, 3, 5, 7, 11, and 15 days after introduction. Upon removal, any beetles or larvae crawling about on the surface of the insulation were brushed off and the samples of each type of insulation from each replicate were placed immediately into individual alcohol baths to halt mealworm activity. Polystyrene and polyurethane panels were sectioned and examined as before. The fiberglass samples were placed on blotting paper and teased apart with foreceps under a stereoscope. The number

and lifestages of A. diaperinus recovered were similiarly recorded. After air drying for four to six hours, the insulation samples (minus infesting organisms) were weighed.

Barke and Davis (1969) demonstrated the applicability of Dyar's law for instar determination, thus validating the use of head capsule widths as indices for establishing larval instar catagories. Larval categories were defined as follows: first instar larvae (pearly-white in color) and second instar larvae (golden-brown in color) were counted as early instars, having head capsule widths of 0.20-0.30mm and 0.35-0.37mm respectively; eighth instar larvae and above were counted as late instars and had head capsule widths ranging from 1.16mm-1.51mm; middle instar larvae constituted all instars (3rd-7th) with head capsule widths between these two ranges.

Results

Study 1. The recovered lifestages of A. diaperinus have been plotted as average numbers on two separate graphs. Figure 2 presents the average number of late instar larvae, pupae, and adults recovered from within the panels over the thirty-five days of the intial test. Figure 3 presents a similiar graph of the eggs and early instar larvae which were, for the most part, recovered from the ethanol stop

baths or from the surface of the panels and may be considered as non-invasive lifestages. Middle instar larvae were rarely recovered from the polystyrene panels and therefore are not included in the figures.

Eggs were usually found deposited singly or in clusters in the cracks between beads of polystyrene. Newly-hatched first instar larvae comprised the bulk of the early instars recovered from the alcohol baths or panel surfaces. Although second instars were recovered, only a few were found to have burrowed into the styrofoam. The majority of styrofoam destruction was attributed to late instar larvae and adults. Tunnels and pupation chambers were littered with small chewed bits of styrofoam, fecal material, and occasionally particles of surrounding media (chicken manure, feathers, ect.).

The general patterns of infestation into polystyrene insulation by these two lifestages of A. diaperinus (late instars and adults) over the thirty-five day test period were compared (Table 1). In the second experiment (Table 2), when fresh panels were introduced into the stock colonies, the same general trend was apparent during the early stages of styrofoam infestation.

The two most abundant arthropods other than A. diaperinus encountered within the tunnels were two species

of mites. The most numerous was a mite (Family: Uropodidae) which was often found in aggregates. The second mite, a smaller unidentified white mite with large chelicerae, appeared singularly except when clustered in small groups about the eggs of A. diaperinus. Other arthropods found occasionally within the galleries included Tribolium spp. adults, and histerid larvae and adults (Carcinops spp.).

Study 2. The average number and lifestages of Alphitobius diaperinus recovered from each type of insulation are presented in Table 3.

The average differences between pre-introduction weight and post-introduction weight of each insulation type are given in Table 4.

Structural damage to the polystyrene panels was in the form of tunnels and burrows, whereas polyurethane panel damage was of a more pitted or pocked nature confined to the panel surface, with tunnels being much less numerous. Fiberglass samples were torn apart by mealworm activity.

Other arthropods consisted of histerid beetles, Tribolium spp. and Uropodid mites and were found most frequently in the fiberglass.

Discussion

Study 1. Figure 3 indicates considerable egg mortality had occurred in the first part of the experiment. Many of the eggs that were counted appeared somewhat dessicated. However, a notable portion did hatch. Although the nutritional value of polystyrene is dubious, many of the newly-hatched larvae survived their first molt as evidenced in the concurrence of second instar larvae with first instar exuviae. The presence of frass, debris, mites, and other possible sources of nutrition within the tunnels may conceivably support further larval development, yet very few third instar larvae were found in or on the panels. This suggests that most of the second instars leave the panels in search of food. Adult A. dipaerinus deposited eggs on the exterior of panels infested with late instar larvae (Days 0-13). But as more adults began to be recovered from the interior of the panels (Days 13-35), oviposition on the exterior of the panels declined. It is interesting to note that at no time were any eggs ever discovered within the tunnels themselves.

The data indicate that adult A. diaperinus do not intiate actual tunneling into the material. As Tables 1 and 2 show, it is the late instar larvae which intiate tunneling, apparently seeking sheltered pupation sites.

Upon entering the insulation, these late instars undergo a three to five day prepupal period before metamorphosing (Figure 2). During this time they remain quiescent. The pupation chambers, lined with chewed bits of insulation, probably represent thermally-stable environments. Yet the average number of pupae recovered from the styrofoam is considerably less than that of the incoming late instar larvae. This indicates that there is mortality occurring at the prepupal and pupal stages. Frequent discoveries of partially-eaten prepupae, pupae, and teneral adults within the tunnels point to a probable cause of such high mortality. Both incoming adults and late instar larvae, by virtue of their association with such cannibalized cadavers, can be implicated in the mortality of pupating and eclosing individuals.

By the twenty-third day post-introduction, most of the panels remaining in the stock colonies had become severely riddled throughout with tunnels. The number of late instar larvae moving from the media and into the panels at this time declined for two related reasons; structural deterioration had depreciated the panels' suitablility as pupational sites, and the rapid influx of adults posed a threat to pupating larvae. As more and more foraging adults moved into the tunnels and expanded them, the presence of all other lifestages (even eggs) decreased.

In a poultry house in which an infestation of polystyrene by A. diaperinus was beginning, late instar larvae would tend to migrate farther and farther up into the structures once a damage threshold or perhaps an adult population density had been reached in other regions of the wall panel. This activity would destroy more and more of thg styrofoam's insulative capacity. Cannibalism, particularly at the very high densities encountered in some deep-pit poultry houses, probably plays a major role in the adaptive behavior of late instar A. diaperinus larvae to migrate into protected areas (such as insulation) that are presumably unattractive to foraging adults. But as more and more tunnels are produced by the late instars, adults begin to follow. The reasons are speculative. Most likely adults, in response to density pressures occurring in the media, are dispersing and, upon entering the tunnels made by the late instars, encounter pupating individuals and consume them. Perhaps adults are following larval tunnels and/or making their own in an active search for food. Adult A. diaperinus are not entering polystyrene insulation in order to lay eggs, even though eggs are deposited upon the surface, because no eggs were found to occur in the tunnels.

Study 2. Polyurethane appears to be favored over polystyrene by ovipositing beetles. Late instar larvae seem

to prefer polystyrene over polyurethane for pupational sites (Table 3). This may be due, in large part, to the different textures of the two materials. The polyurethane is much more crisp and abrasive than the softer, more pliant polystrene, tending to discourage tunneling. Being an open-cell formulation with no filler material, the polyurethane had more crevices on its surface for oviposition than does the open-celled polystyrene with a filler matrix. The filler material fills in the interstitial spaces in the polystyrene, making its surface smooth. The polystyrene was soft enough, however, that adult beetles were observed to penetrate the material with their ovipositors and deposit eggs just beneath the surface. These findings should not be misconstrued to imply that A. diaperinus larvae will bore into polystyrene but not polyurethane. It has been shown that late instar larvae will readily bore into both polystyrene and polyurethane (Ichinose, et al. 1980) and infestations have been observed in both materials on various Virginia poultry houses. The results suggest only that, when given a preference, polystyrene will be utilized by late instar larvae before polyurethane, and that tunneling behavior may be a function of material texture more than anything else.

Fiberglass insulation, when infested with Alphitobius diaperinus is not employed as pupational sites and all lifestages, particularly adults and middle instar larvae, were found in the fiberglass samples (Table 3). Very few pupae were found in the fiberglass samples.

The average weights of both polystyrene and polyurethane were progressively depleted as the result of infestation, whereas the fiberglass samples became heavier (Table 4), undoubtedly due to the increase of foreign material (frass, media, etc.) that was brought into the fiberglass by the invading insects. In all cases, exposure of the materials to A. diaperinus probably has a deleterious effect on the R-value (insulative capacity) of these materials.

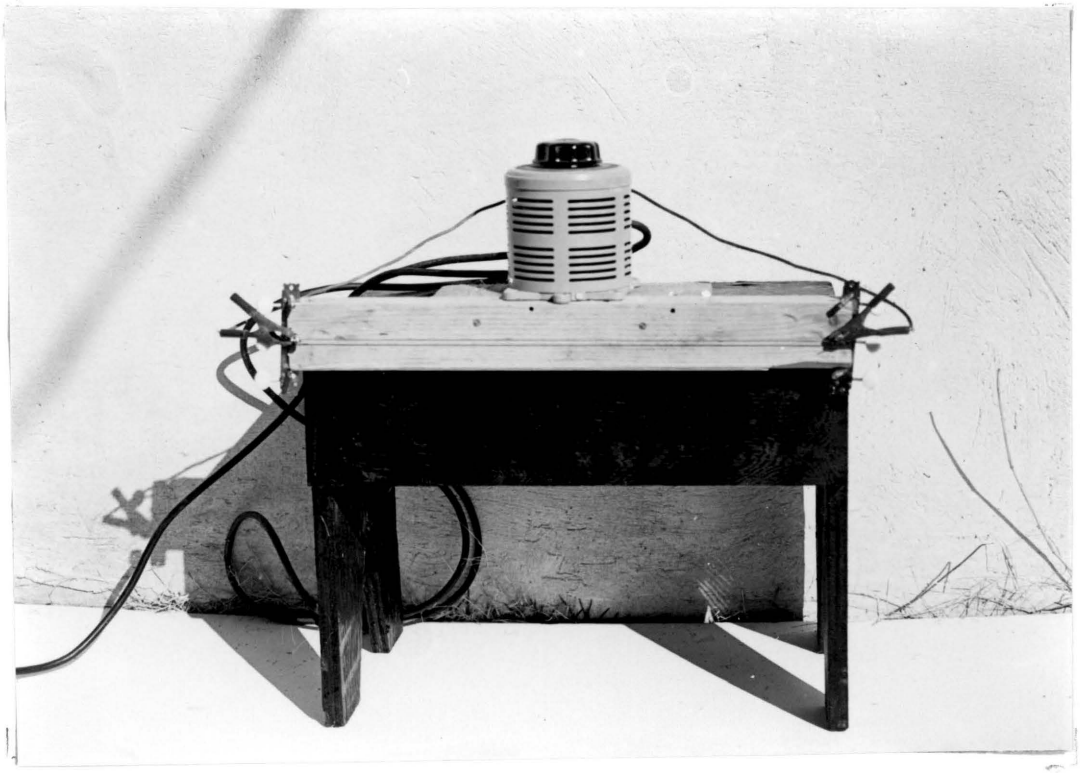

Figure 1. Styrofoam sectioning apparatus.

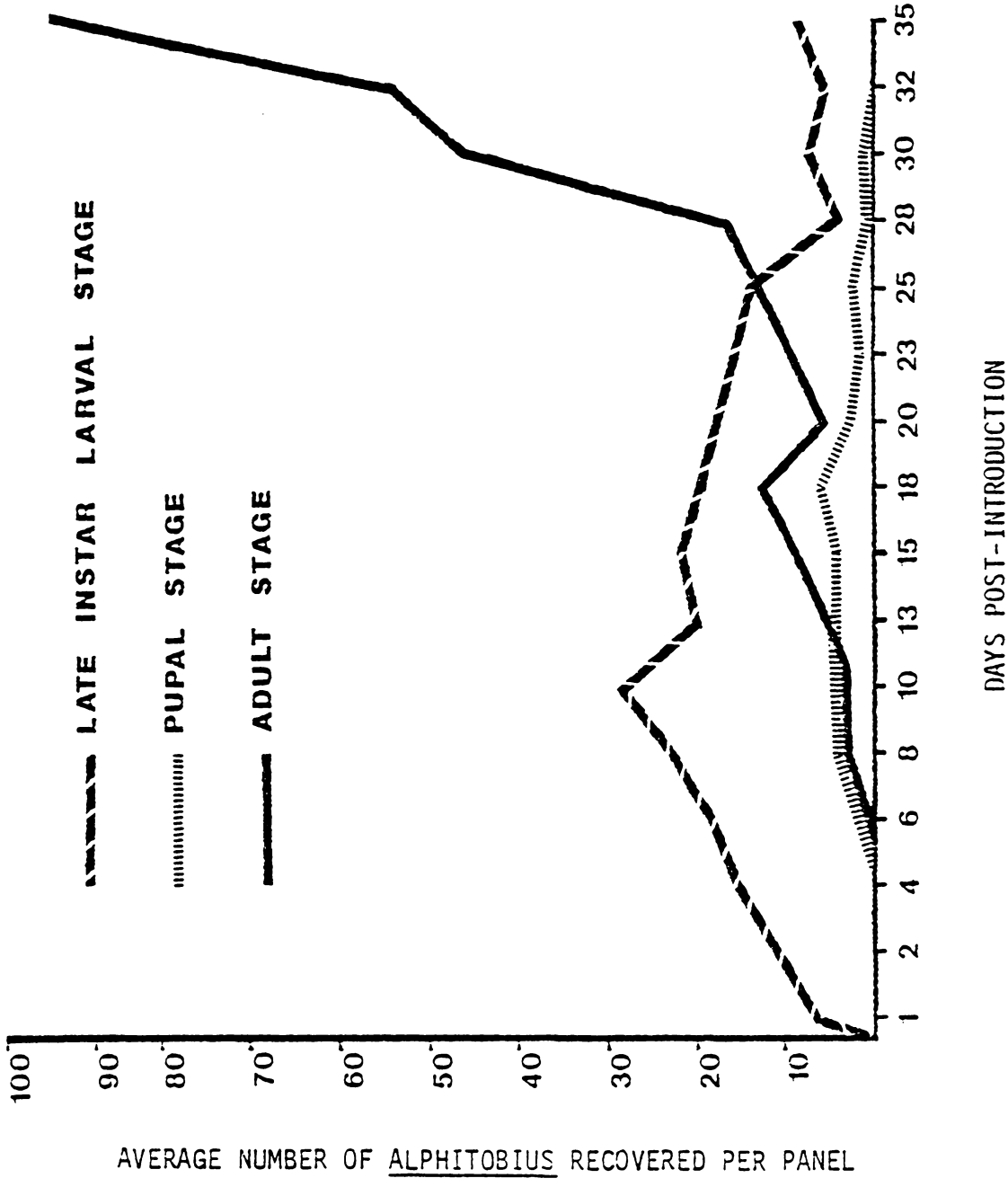

Figure 2. Average number of Alphitobius diaperinus recovered from polystyrene panels (panel volume=approx. 52cm^3). Adult, late instar larval and pupal stages.

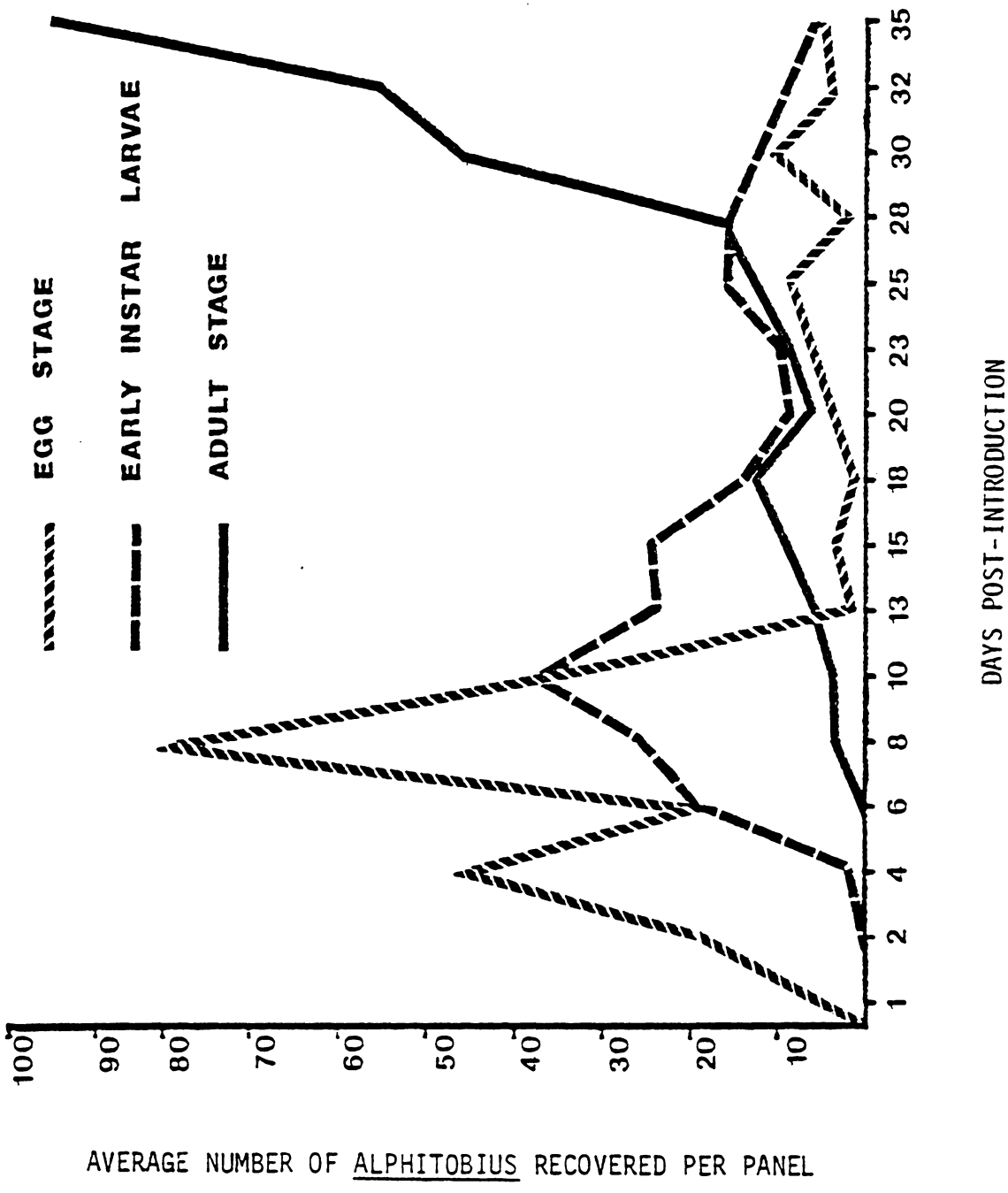

Figure 3. Average number of Alphitobius diaperinus recovered from polystyrene panels (panel volume=approx. 52cm^3). Egg, early instar larval and adult stages.

Table 1. Number and percentage of late instar (LL), pupal (P), and adult (A) Alphitobius diaperinus recovered from polystyrene panels. (Intial test).

DAY POST-INTRODUCTION	%LL	%P	%A	TOTAL NUMBER RECOVERED
2	100.00	0.00	0.00	54
4	98.55	1.45	2.53	69
6	93.55	3.23	3.23	93
8	78.47	11.11	10.42	144
10	80.45	11.17	8.38	179
13	71.94	10.07	17.99	139
15	61.49	12.85	25.70	179
18	52.88	14.14	32.98	191
20	68.66	8.96	22.39	134
23	60.31	7.63	32.06	131
25	47.02	7.95	45.03	151
28	18.70	4.07	77.24	123
30	14.13	2.17	83.70	276
32	14.15	0.64	89.07	311
35	8.25	0.00	91.75	521

Table 2. Number and percentage of late instar (LL), pupal (P), and adult (A) Alphitobius diaperinus recovered from polystyrene panels. Second test started one week after completion of intial test.

DAY POST-INTRODUCTION	%LL	%P	%A	TOTAL NUMBER RECOVERED
1	100.00	0.00	0.00	34
2	100.00	0.00	0.00	60
4	93.26	2.25	4.49	89
6	77.05	10.66	12.30	122

Table 3. Average number and lifestage[1] of Alphitobius diaperinus recovered from samples of three insulation types.

DAY POST-INTRODUCTION	POLYSTYRENE						POLYURETHANE						FIBERGLASS					
	E	EL	ML	LL	P	A	E	LL	ML	LL	P	A	E	LL	ML	LL	P	A
1	0.0	0.0	0.0	0.0	0.0	0.0	19.7	0.0	0.0	0.3	0.0	0.0	48.7	1.3	88.0	17.7	0.0	45.3
3	0.0	0.0	0.0	12.3	0.0	3.0	86.3	0.0	0.0	0.0	0.0	0.0	57.7	0.7	136.7	109.0	0.0	202.3
5	0.0	0.0	0.0	19.0	0.0	4.3	24.0	20.0	0.0	0.3	0.0	1.0	48.6	79.0	116.7	47.3	0.0	161.0
7	0.0	1.0	1.7	14.3	0.0	31.0	4.0	5.0	0.0	2.3	0.0	5.0	13.3	34.6	131.7	52.3	0.0	206.0
11	0.0	0.0	0.0	4.7	0.0	7.7	2.0	1.0	0.3	3.3	0.0	4.3	19.3	11.7	76.7	37.0	0.0	190.3
15	0.3	0.0	0.3	1.7	0.0	4.3	0.0	0.0	0.0	0.0	0.0	13.7	12.3	12.7	32.3	19.0	0.0	145.0

[1]E=egg stage, EL=early instar larval stage, ML=middle instar larval stage, LL=late instar larval stage, P=pupal stage, A=adult stage.

Table 4. Average weight change (grams)[1] in three insulation types following infestation by Alphitobius diaperinus.

DAY POST-INTRODUCTION	POLY-STYRENE	POLY-URETHANE	FIBERGLASS
1	+0.03	-0.26	+0.08
3	+0.07	-0.07	+0.35
5	-0.17	-0.13	+0.83
7	-0.27	-0.30	+0.57
11	-0.52	-0.57	+0.85
15	-0.57	-0.68	+1.28

[1]Average pre-introduction weights of the three insulation types were: polystyrene 4.10 ± 0.51g.; polyurethane 3.14 ± 0.45g.; fiberglass 5.96 ± 0.38g.

IV. FEEDING BEHAVIOR AND OTHER OBSERVATIONS

Introduction

Although A. diaperinus is one of the most abundant arthropods found in poultry litter and manure, and found to be quite voracious, little work has been published on its potential as a predator of the housefly, Musca domestica L. Toyama and Ikeda (1976) reported it feeding only on first instar house fly larvae. Koszlov (1970) found A. diaperinus to be an effective predator of the chicken mite Dermanyssus gallinae (DeGreer). A closely-related species, Alphitobius laevigatus (Fab.), has been reported as a predator of flea larvae (Fox and Bayona 1968). In casual observations, A. diaperinus adults and larvae have been observed attacking and feeding upon larvae and pupae of house flies when put together in a Petri dish.

House fly larvae display optimal growth in chicken manure at a temperature range of 34-38°C and a substrate relative humidity (RH) of 70% (Teotia and Miller 1973). Third instar larvae migrate to drier substrates to pupate. Preliminary measurements taken in the pit of a high-rise caged layer house in Amelia Co., VA indicate that adult A. diaperinus are most abundant at a substrate RH of about 53%.

The substrate temperature under the birds was found to range from 32°- 36°C. A. diaperinus apparently inhabits drier substrate conditions than first and second instar housefly larvae. Thus house fly predation by A. diaperinus is most likely to involve late instar, pupal and eclosing house flies, for it is at these stages in the life history of the house fly where there would be increased probability of the two species (i.e. Musca domestica and A. diaperinus) coming in contact with one another.

Cannibalism by A. diaperinus has been reported in laboratory colonies (Barke and Davis 1969, Sarin 1978). Saxena and Sarin (1971) reported chitinase activity in the digestive juice of A. diaperinus larvae but not in adults. They stated that only larvae are cannibalistic. I suspected that adult A. diaperinus were also cannibalistic and that cannibalism, particularly of prepupae and pupae, may be an important factor in causing late instar larvae to tunnel into insulation, seeking protected pupational sites.

Objectives

The objectives of the reseach presented here were 1) to determine the potential of A. diaperinus as a predator of metamorphosing house flies and 2) to determine which lifestages of A. diaperinus are cannibalistic. In addition, observations on feeding and flight activity were recorded.

Materials and Methods

House flies used in the predation tests were from the V.P.I. & S.U. colony.

Flytest 1. Predation of pupal and eclosing house flies by A. diaperinus was examined. Nine, open-lid, half-pint Mason jars containing fifty house fly pupae each were placed into cylindrical half-gallon ice cream cartons. Five A. diaperinus were added to three of the Mason jars. Three other Mason jars recieved 5 middle instar larvae of A. diaperinus. The remaining three jars served as controls. The ice cream cartons were fitted with emergence tops (Figure 4). The cages were held at room temperature (ca. 21°C) and the number of flies that had emerged were counted at the end of 2 weeks.

The cage design was such that any flies that had successfully emerged and hardened could take flight to avoid predation and would not necessarily be confined to the same container as A. diaperinus. This was done to alleviate the difficulty in determining whether reduction in the numbers of house flies was the result of actual predation on eclosing flies or merely the result of scavenging after the flies had died.

Flytest 2. Predation of prepupal and pupating house fly larvae by A. diaperinus was examined. Twenty late instar

house fly larvae were placed into twelve half-pint Mason jars fitted with special emergence tops (Figure 5). Five A. diaperinus adults were placed in four of these cages, five A. diaperinus middle instars were placed in another four and the remaining four cages served as controls. The jars were held in an environmental chamber at 27°C for two weeks and examined periodically. Any flies in the emergence chambers were collected and put into vials containing 70% ethyl alcohol. At the end of two weeks the jars were emptied and the number of intact flies and the number of opened and unopened pupal cases were recorded.

Flytest 3. The third investigation was designed to simulate natural conditions in the pit of a high-rise caged layer house and to test the effects of a small population (ratio of 1 beetle to 4 house fly larvae) of A. diaperinus on adult house fly emergence.

Chicken manure was collected from caged layers at the Poultry Science Dept., VPI & SU, and frozen for three days to sterilize the manure. Then the manure was thawed, warmed in a water bath, and placed into 7 plastic-lined pint cups which were then seeded with 100 first instar house fly larvae. The seeded manure containers were placed into cylindrical half gallon cartons. The intervening space between the manure cups and the wall of the outer container

was filled to within 1/2 - 1 in. of the level of the manure cup with coarse sand. A mixture of sterilized 16% protein standard layer diet and split fresh green corn kernels (for moisture) was spread over the coarse sand to the level of the manure cup. This represented a drier environment and provided pupational sites for the developing fly larvae (Figure 7). Twenty-five adult A. diaperinus were placed into four of these containers. Three of the containers served as controls. The containers were placed in a controlled environment chamber at 27°C at 24 h darkness. All manure cups were lightly misted with distilled water every other day to prevent the manure from drying out and to ensure maggot migration into the surrounding habitat at time of pupation. Ten days later the emerged flies were collected and counted. At the end of two weeks the colonies were torn apart and the number of whole flies and the number of opened and unopened pupae were counted and recorded.

Data Analysis. Raw data for Flytest 1 were comprised of the number of flies emerged. Raw data for Flytests 2 and 3 were comprised of the number of flies emerged, the number of unopened puparia recovered (an indication of mortality due to natural causes), the number of normally-opened puparia recovered (an indication of the number of flies that had successfully pupated), and the numbers of flies that

were presumed to have emerged successfully from their puparia yet were not accounted for. This last category of data was derived by subtracting the total numbers of flies recovered from the total numbers of normally-opened puparia recovered. Each catagory of data was analyzed separately with single classification analysis of variance (ANOVA). Those tests in which a significant difference between groups was indicated were further analyzed to detect which means differed using the Student-Newman-Keuls procedure (SNKP), an a posteriori (unplanned) comparision method using the studentized range to measure differences among means (Sokal and Rohlf 1969).

Cannibalism and other observations. Intact A. diaperinus pupae were placed in glass dishes (ca. 10cm diam.) with three different ratios (5:1, 10:1 25:1) of; adult to pupae, late instar larvae to pupae and early instar larvae to pupae. Adults, larvae and pupae used in this test were from the same stock colony. A. diaperinus pupae were never abundant in the stock colonies and were intially gathered by placing late instar larvae upon moist sand and waiting for them to pupate as described by Preiss and Davidson (1966). However, it was found more expedient to place polystyrene panels of various dimensions into the stock colonies and remove them three or four days later.

The pupae and prepupae which, as late instars, had tunneled into the polystyrene could easily be located by holding the thin panel up to a light source. They were then gently removed with foreceps. The dishes were kept in a growth chamber at 27°C and 24 hr darkness. The test was conducted for the length of the pupal period (4-7 days). Daily observations were recorded.

Observations were made on a colony of A. diaperinus collected from a turkey house in Hardy Co., W. Va. The colony was housed in a 19 liter (5-gallon) glass aquarium filled with ca. 7.5cm of the same broiler litter in which they were collected. The aquarium was placed next to a north-facing window. A small panel of 5cm (2") closed-cell polystyrene set in a wood frame was placed standing up in the aquarium. Observations were noted from March to July.

Results and Discussion

The results for Flytests 1, 2, and 3 are given in Tables 5, 6, and 7 respectively. The results of the cannibalism test are given in Table 8.

Flytest 1. There was significant reduction (alpha=0.05) in the number of house flies emerging in both groups containing mealworms (Table 5). In addition there was higher fly mortality in the cages which contained adult

A. diaperinus (Group 1) as compared with those that contained middle instar larval A. diaperinus (Group 2). These results indicate that adult A. diaperinus are better able to kill and consume house fly pupae and eclosing adults than are middle instar A. diaperinus.

Flytest 2. Results confirm that adult mealworms are more predatory than larval mealworms (Table 6). The only group in which the mean numbers of flies emerged was significantly different at the 0.05 level from that of the control group (flies only) was the group which contained A. diaperinus adults (Group 1). The group containing middle instar mealworms (Group 2) did not show a statistically significant (alpha=0.05) reduction in house fly emergence, although some reduction did occur (Column 1). Column 2 shows the averages of the total number of fly puparia (opened and unopened) recovered from the three groups of cages. This provides an indication of the stage at which house fly mortality within Group 1 occurred. It is apparent that many of the third instar maggots never reached the pupal stage or, if they did, were consumed before the puparium hardened. Once the maggots had successfully pupated, there were no significant differences between groups in mortality during or upon completion of the pupal stage (Columns 3, 4, and 5)). Flytest 2 indicates that

adult A. diaperinus is more predaceous and attacks the prepupal/early pupal stage of the house fly with greater frequency than it does pupal or eclosing adult house flies.

Flytest 3. Preliminary tests (Flytests 1 and 2) demonstrate that A. diaperinus adults cause a significant decrease in the number of adult house flies emerging when placed in the same container. However this has little bearing on what really goes on in the manure of a poultry operation. Flytest 3 attempted to simulate natural conditions.

Results for Flytest 3 (Table 6) show no significant differences at the 0.05 level between the group containing A. diaperinus and the group containing only flies for any of the parameters tested. Although not statistically significant, reduction in house fly emergence (Column 1) and increase in the numbers of flies unaccounted for (Column 4) due to the presence of A. diaperinus did occur. However, the design of the cage (Figure 7) did not prevent A. diaperinus from scavenging on flies that died before they could move up into the emergence chamber. Thus, reduction in the number of house flies due to scavenging by A. diaperinus is difficult to distinguish from reduction due to actual predation.

Under simulated natural conditions at low population densities, A. diaperinus may not be an effective predator of the house fly. Perhaps when given a choice of food items (in this case cracked corn and poultry feed), A. diaperinus will not attack pupating house flies in lieu of alternate food sources. Because of the enormous numbers of A. diaperinus present in the manure of many Virginia poultry houses however, detailed experiments involving various population densities, varying ratios of predator-to-prey, and predation studies under field conditions would be useful. In addition, the role of A. diaperinus, in modification of the manure habitat for house fly control is uncertain at this time.

Cannibalism and Other Observations. Results of the cannibalism experiment (Table 8) show that all lifestages cannibalized pupae. On day 4, pupae were not replaced because the available supply had been exhausted. It was also noted that cannibalism was not restricted only to pupae. Many adults and late instar larvae that had died during the course of the test period had legs missing. Early instar larvae were observed to feed upon the posterior edge of the elytra of a newly-emerged adult. Often adults and larvae used in the test were found partially eaten or were never accounted for. Although both late instar and

early instar larvae molted during the test period, only the early instars were observed to feed upon the exuviae.

When adult A. diaperinus, which had had their elytra removed or had otherwise been mutilated, were placed into a porcelain tray with other adults the other adults would, one-by-one, investigate the wound. Within minutes, one or two adults would begin to feed on the hemolymph and wounded tissue. Once feeding had commenced by just a few individuals, many nearby adults quickly joined the feeding. After ca. 20 minutes, all that would remain of the cannibalized individual would be the elytra and portions of the sterna. Similiar attacks by adult beetles were noted to occur when late instar house fly larvae and dewinged flies and honeybees where added to the tray. These observations indicate that adult A. diaperinus is a voracious omnivore, cannibalistic and predaceous, and probably an efficient scavenger in the poultry house environment.

Although Preiss (1966) found A. diaperinus reluctant to fly, there have been several reports concerning flight activity, most notably when poultry houses have been cleaned out (Thornberry 1978, Jerrard 1980, Smith 1981). Observations on flight activity have been made on a colony of A. diaperinus housed in a glass aquarium and kept in a north-facing window. For 2-3 weeks in mid-June, 1979,

always 1 or 2 hours before sunset, adults would emerge from the litter and begin increased activity. Frequent mating, with coupling lasting 30-45 sec., took place at this time. Many beetles climbed up the wood and styrofoam structures within the aquarium, spread their elytra for a moment, then flew into the glass facing the window. This activity occurred on a regular basis for ca. 2 weeks but became less and less frequent, finally ceasing as the summer progressed. Since this behavior occurred only on cloudless evenings it is presumed that there may be some sort of photoperiod response at work. Similiar activity of A. diaperinus colonies were noted to occur during the early summer of the subsequent two years (1980-81). Preliminary investigations could not demonstrate a marked phototropism by adult beetles to either white or ultraviolet light.

Adult flight may be one mechanism by which A. diaperinus colonizes the manure or litter of poultry operations that have just been built. It is known that A. diaperinus exists in the wild, associated mostly with wild birds or bats, and these natural populations may serve as reservoirs for infesting newly-built poultry houses. Alphitobius diaperinus is also known to be a pest of stored grain, existing in the warehouses of feed mills. Thus, this insect may also gain access to newly-constructed poultry houses by way of the poultry feed.

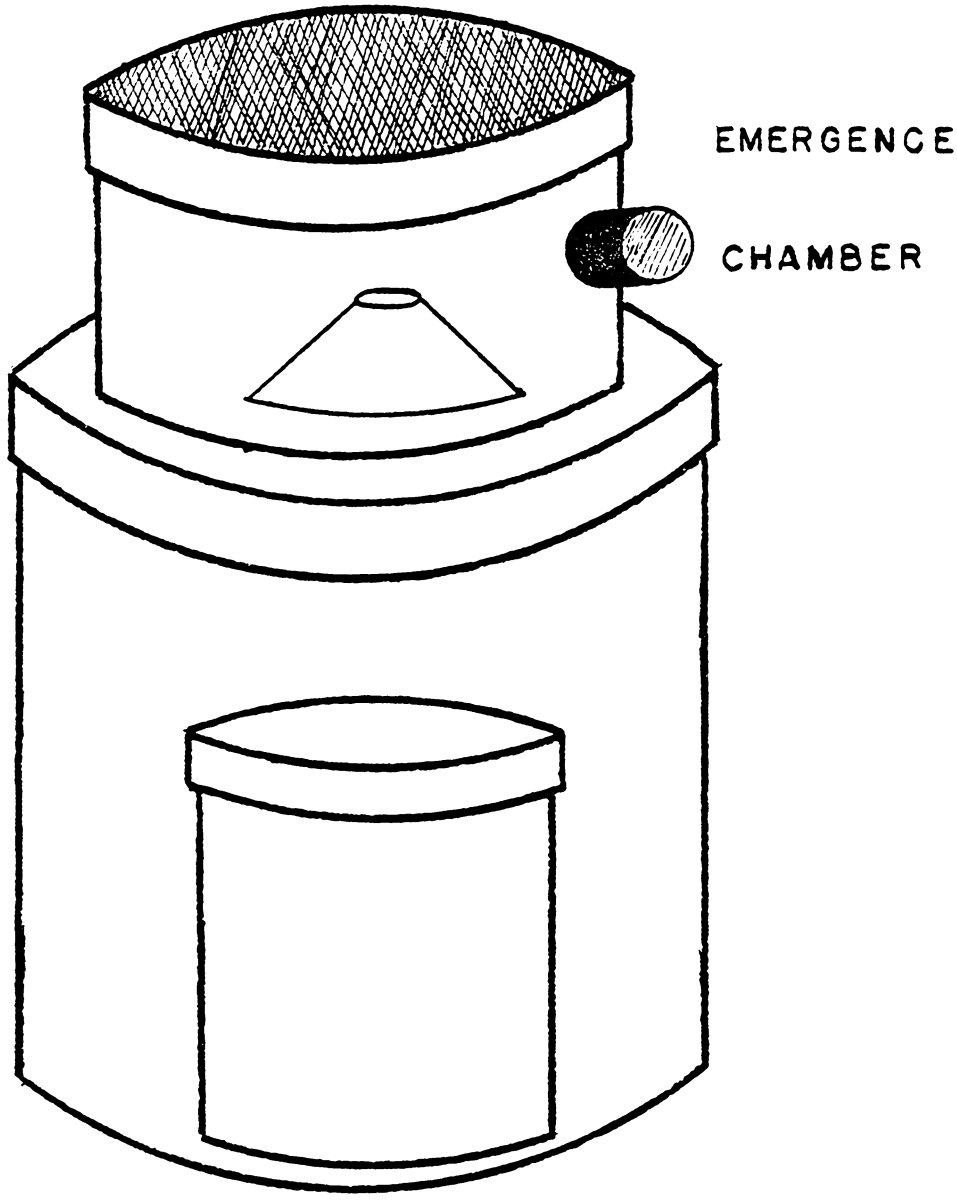

Figure 4. Emergence cage used in house fly predation test. Flytest 1.

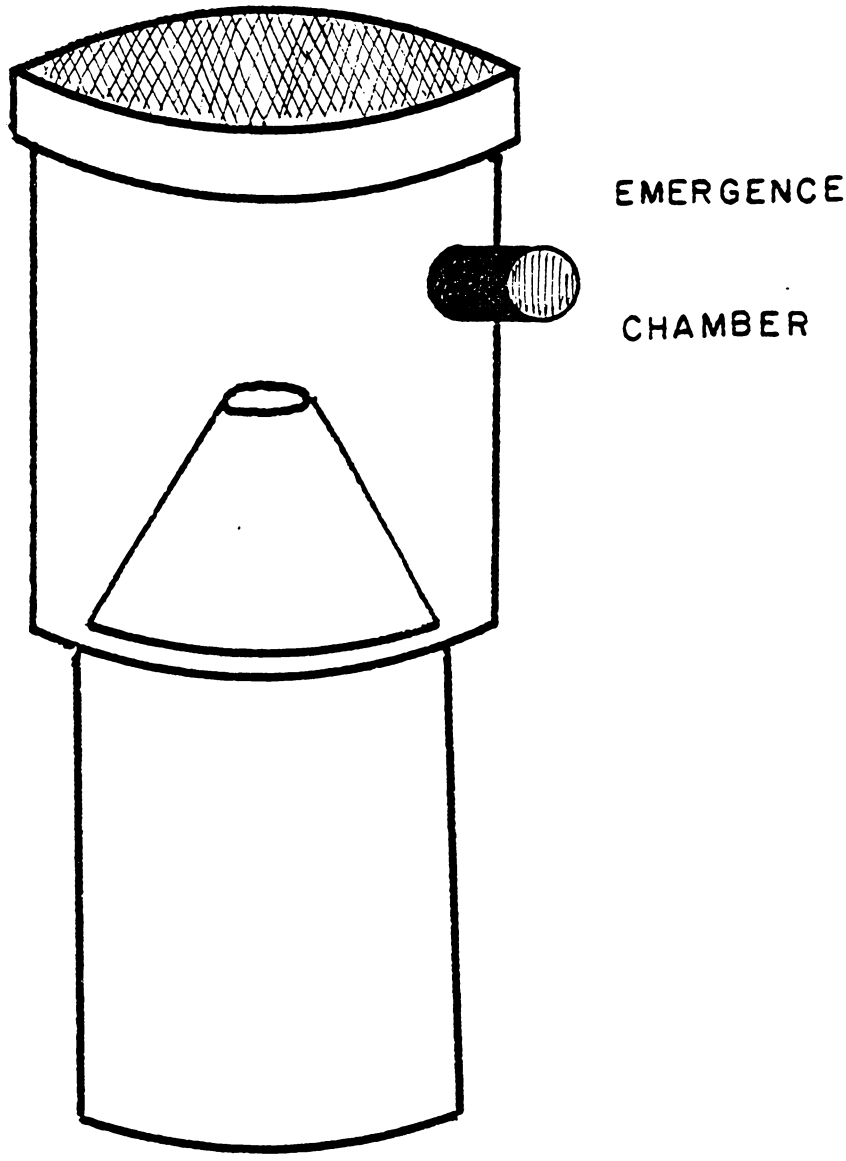

Figure 5. Emergence cage used in house fly predation test. Flytest 2.

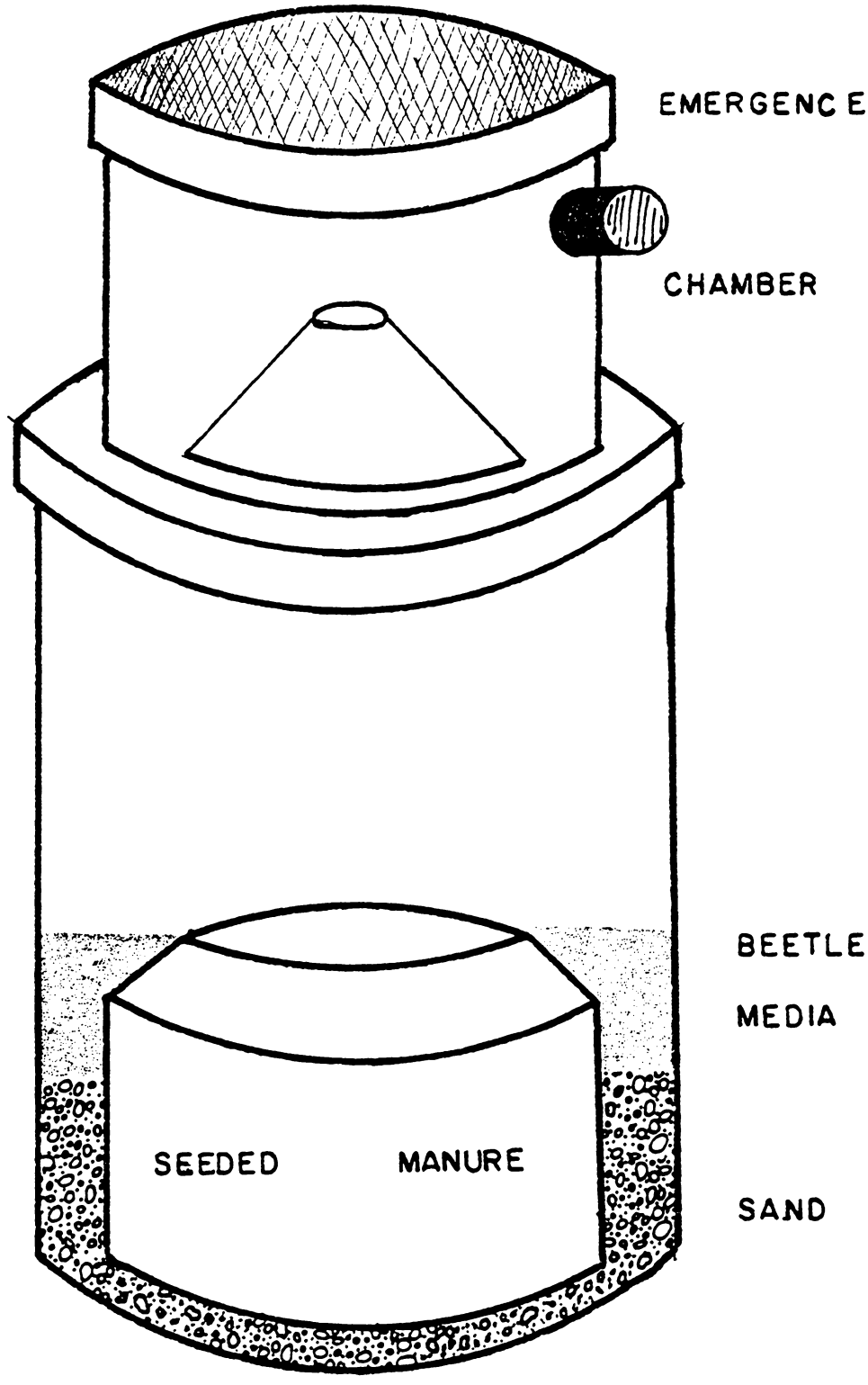

Figure 6. Emergence cage used in house fly predation test. Flytest 3.

Table 5. The effect of adult and larval Alphitobius diaperinus on house fly adult emergence under laboratory conditions. Cells contained 5 A. diaperinus and 50 house fly pupae. Flytest 1.

	GROUPS	MEAN NO. FLIES EMERGED[1]
1.	Flies + A. diaperinus adults	31.33
2.	Flies + A. diaperinus larvae	36.33
3.	Control--- Flies only	47.00

[1]all means significantly different using Student-Newman-Keuls test at an alpha level of 0.05.

Table 6. The effect of adult and larval Alphitobius diaperinus on house fly adult emergence under laboratory conditions. Cells contained 5 A. diaperinus and 20 late instar house fly larvae. Flytest 2.

GROUPS	MEAN NO. FLIES EMERGED[1]	MEAN NO. PUPARIA RECOVERED	MEAN NO. UNOPENED PUPARIA RECOVERED	MEAN NO. OPENED PUPARIA RECOVERED	MEAN NO. FLIES PRESUMED EMERGED BUT NOT ACCOUNTED FOR
1. Flies + A. diaperinus adults	7.00b	15.75b	5.50a	10.25a	3.25a
2. Flies + A. diaperinus larvae	9.50ab	17.00ab	4.25a	12.75a	3.25a
3. Control-- flies only	12.75a	19.00a	6.25a	12.75a	3.25a

[1]means within columns with same letter are not significantly different at the 0.05 level.

Table 7. The effect of adult Alphitobius diaperinus on house fly adult emergence under simulated poultry house conditions. Cells contained 25 A. diaperinus adults and 100 first instar house fly larvae. Flytest 3.

GROUPS	MEAN NO. FLIES RECOVERED	MEAN NO. UNOPENED PUPARIA RECOVERED	MEAN NO. OPENED PUPARIA RECOVERED	MEAN NO. FLIES PRESUMED EMERGED BUT NOT ACCOUNTED FOR
Flies + A. diaperinus	48.50	2.25	73.00	24.50
Flies only (Control)	67.67	1.33	74.00	6.67

[1]means within columns are not significantly different at the 0.05 level.

Table 8. Incidence of cannibalism (X) of pupal (P) Alphitobius diaperinus by adult (A), late instar larval (LL), and early instar larval (EL) Alphitobius diaperinus.

	DAY POST-INTRODUCTION			
RATIO	1	2	3	4
A:P				
5:1				X
10:1	X	X	X	No Pupae Available
25:1			X	"
LL:P				
5:1			X	"
10:1		X	X	"
25:1	X	X	X	"
EL:P				
5:1			X	"
10:1		X	X	"
25:1			X	"

V. INSECTICIDE STUDIES

Introduction

The choice of insecticides which are approved for use in Virginia poultry operations are limited because of the possibility of insecticide residues occurring in the meat and eggs of poultry. Three of the most commonly used chemicals are tetrachlorvinphos, carbaryl, and permethrin. Tetrachlorvinphos is presently approved for use in Virginia as a direct spray on caged layers, a premise spray, and a manure and litter treatment. Both carbaryl and permethrin are approved for use as a direct spray on caged layers, a premise spray, and a litter treatment. They are not approved for use as a manure treatment (Roberts 1982). At the present time there are no control recommendations against the lesser mealworm, A. diaperinus.

Objectives

The objectives of this study were to investigate the effectiveness of tetrachlorvinphos, carbaryl, and permethrin against adult A. diaperinus 1) on treated polystyrene insulation, 2) on treated plywood, 3) late instar larval A. diaperinus on treated polystyrene insulation and 4) to

assess the comparative toxicities of each compound by topical applications of the three insecticides using adult A. diaperinus.

Materials and Methods

The insecticides used were: the organo-phosphate insecticide, tetrachlorvinphos or Rabon (2-chloro-1 (2,4,5-trichlorophenyl)-vinyl dimethylphosphate); the carbamate insecticide, carbaryl or Sevin (1-naphthyl N-methylcarbamate); and the synthetic pyrethroid insecticide, permethrin or Ectiban (3-(phenoxyphenol) methyl (+) cis, trans-3-(2,2-dichloroethenyl)-2,2-dimethyl cyclopropane carboxylate).

Panel tests. In Test 1, square foot panels of one inch closed-cell beaded polystyrene were sprayed on one side with three different dosages (0.1%, 0.05%, and 0.01% a.i.) of permethrin 25% WP, permethrin 5% EC, and carbaryl 80% WP. Tetrachlorvinphos 50% WP (0.50% a.i.) was used as a standard. The appropriate amount of each insecticide was measured on a Meittler balance and mixed with 2ml distilled water in a 5ml test tube. Three replicates of each dose and three control tubes (distilled water only) were prepared as described and sprayed on the surface of the polystyrene panels with a chromotography sprayer. The sprayer was

positioned about a foot away from the surface to be sprayed and the insecticide was applied as evenly as possible over the entire square foot area. The panels were allowed to age overnight. After 24 h, ten adult A. diaperinus were retained upon the surface of each panel for one hour. To prevent escape the beetles were confined beneath large (14cm in diam.) overturned Petri dishes held in place by rubberbands (Figure 7). After one hour exposure, the beetles were removed and placed in clean half-pint Mason jars. The recovery jars were held at room temperature (ca. 21°C) and mortality counts were taken at 24 and 48 h post-exposure. Death was defined to be the inability to crawl when prodded with forceps. Because of their tendency to feign death, the mortality counts were taken only after the beetles had been placed in a dissecting tray that had been heated such that it was warm to the touch.

Exposure to the treated surfaces and subsequent 24 and 48 h mortality counts were performed on a weekly basis for 21 weeks. Recovery jars were held at room temperature and thoroughly washed at the end of each weeks' test.

Similiar tests were performed with adult A. diaperinus on treated square foot interior grade plywood panels (Test 2) and with late instar A. diaperinus larvae on square foot polystyrene panels (Test 3) using the same three dosage

levels of permethrin 25% WP and carbaryl 80% WP. Tetrachlorvinphos 50% WP (0.5% a.i.) was used as a standard. Permethrin 5% EC was excluded from these tests. Only five larvae were used per replicate in Test 3.

Topical Applications. Six doses of each insecticide (tetrachlorvinphos, carbaryl, and permethrin) were applied topically to the prosternum of adult beetles. Measured drops of insecticide were applied with an ISCO microapplicator (Instrument Specialitiies Co. Inc., Lincoln, Neb.) through a 1/4 cc Tuberculin syringe (Beckton-Dickinson, Rutherford. NJ). The delivery rate of the syringe was calibrated gravometrically. The delivery rate was 0.6 microliter per second and the delivery time used for each application was one second, making the volume of liquid applied per beetle 0.6 microliter.

Preliminary topical applications were conducted to determine appropriate dosage ranges. Six dosages were used for tetrachlorvinphos and carbaryl. They ranged from 10.00 mg/ml (6.00 ug/beetle) to 0.03 mg/ml (0.018 ug/beetle). The six dosages used for permethrin ranged from 1.00 mg/ml (0.60 ug/beetle) to 0.003 mg/ml (0.0018 ug/beetle). A control group was run with each insecticide type and each dosage of each insecticide was replicated three times.

Twenty beetles were treated for each replicate. Age and sex were not standardized. However, since it is known that female A. diaperinus are generally larger in size than the males, only larger individuals which appeared healthy and vigorous were selected for testing. Beetles were not anesthetized and were handled uniformly by grasping about the lateral edges of the elytra with forceps. Insecticide was applied on the prosternum of the beetles whereupon the drop spread rapidly into adjacent cuticular folds. Treated beetles were placed in clean half-pint Mason jars and held at 27°C in a controlled environment chamber. A 24 h mortality count was taken, using the same criteria of death as described in the panel tests.

Observations on the behavioral toxic effects of each compound on ten A. diaperinus adults were recorded. An intermediate dose of each compound (as determined from the results of the topical applications) was applied as before. These were; permethrin 0.1 ug/ul, tetrachlorvinphos 0.3 ug/ul, and carbaryl 1.0 ug/ul, plus a control (acetone only). Beetles were placed in a large watchglass for observations and maintained at 27°C in an environmental cabinet. Observations were recorded immediately upon treatment, at ca. 15-minute intervals for the first hour, ca. 30-minute intervals for the second hour, and once an

hour for the following four hours post-treatment. A final observation was recorded 24 h post-treatment.

Data Analysis. Dose-response curves for the topical applications were computed by the probit procedure included in the Statistical Analysis System (SAS) software package. Since no mortality occurred in the control groups, it was not necessary to use Abbott's formula (Abbott 1925) to correct the percentage mortality.

In the panel tests, the weekly percent mortalities at each dosage level were corrected for mortality that occurred in the controls using Abbott's formula. The corrected percent mortalities for each panel test were first analyzed by factorial analysis of variance (ANOVA) with treatments, week, replicate, and hour (24 h vs 48 h mortality counts) as the main effects. This preliminary ANOVA was performed to detect possible discrepancies between replicates (replicate effect), and to detect any possible recovery from toxic effects that the insects might have exhibited between the 24 h and 48 h counts (hour effect).

When no hour or replicate effect were evident, the data for each treatment per week were pooled. Separate ANOVA's on each week were performed to generate the degrees of freedom (df) and error mean squares (MS) for each week to be used in the Duncan's Multiple Range Test (DMRT). Separate

DMRT's were performed on each week's data to test for differences between the mean corrected percent mortalities due to treatments.

When there was a significant hour effect (Test 1), the treatments were partitioned according to hour (24 vs 48), creating a new variable 'trour'. A DMRT, using the df and MS generated from the preliminary ANOVA, was performed on the variable 'trour' to test for significant differences between the mean corrected percent mortalities. In this way the treatments which caused the significant hour effect in the preliminary ANOVA could be identified. Separate DMRT's were performed on each week's data as described before, with the exception that only the mortalities from the 48 h count were included in the analysis.

Results

Panel Tests. None of the three panel tests showed a significant replicate effect at the 0.01 alpha level. Test 1 (adult beetles on treated styrofoam) was the only panel test which exhibited a significant hour effect (alpha=0.0001). Although all treatments showed slight recovery between the 24 h and 48 h counts, a DMRT on the partitioned treatments shows that this recovery was only statistically significant (alpha=0.05) with adult A.

diaperinus exposure to styrofoam panels treated with the higher dosages of permethrin EC and with the lowest dosage of permethrin WP (Table 9).

Table 10 shows the results of Test 1. Since effective control can only be regarded as those individuals which are actually killed by a particular treatment and not merely knocked down, Table 10 presents only the mean mortalities from the 48 h count. Tables 11 and 12 show the results of Tests 2 and 3 respectively. Since there was no significant hour effect in these tests, the data presented represent averages of both 24 h and 48 h mortality counts.

In Test 1 (Table 10), the emusifiable concentrate formulation of permethrin at all doses was found to be less effective against adult A. diaperinus than the wettable powder formulation of permethrin. At the highest dose (0.10% a.i.), permethrin EC was only effective (mortality > 75%) after one week. Permethrin WP and carbaryl WP at dosages of 0.10% a.i. and 0.05% a.i. appeared to have been comparable in their residual activity up to eight weeks post-spray. At the lowest dose (0.01% a.i.), carbaryl appeared to be slightly more effective than permethrin intially. Permethrin WP at 0.10% a.i. and tetrachlorvinphos WP at 0.50% a.i. maintained an extremely high degree of residual activity (mortality > 90%) for the entire 21-week

test period. Since permethrin EC was not as effective as permethrin WP in Test 1, it was not included in subsequent panel tests.

Table 11 presents the results of Test 2. The residual activity of carbaryl WP on plywood at all dosges tested was comparable with than that of permethrin WP. Neither compound at the low dose (0.01% a.i.) or the intermediate dose (0.05% a.i.) produced mortalities in excess of 75% after the first week. At the highest dose tested (0.10% a.i.), carbaryl WP produced mortalities in excess of 75% for five weeks post-treatment whereas permethrin WP at the same dose lasted only three weeks. Tetrachlorvinphos WP at 0.50% a.i. provided 100% control for the entire 11-week test period.

Table 12 presents the results of Test 3. Intially the activity of carbaryl WP at the highest dosages (0.10% a.i. and 0.05% a.i.) was comparable with that of permethrin WP at the same dosage levels and both showed a high degree of control (mortality > 90%). After the seventh week post-spray, carbaryl WP was significantly less effective than permethrin which consistently produced higher percent mortalities until the eighteenth week. At the lower dosage (0.01% a.i.) both permethrin and carbaryl were ineffective (mortality < 75%) after the third week. Tetrachlorvinphos

WP at 0.50% a.i. provided excellent control (mortality > 80%) for the entire 21-week test period. Larval molting, accompanied by the cannibalism of the newly-emerged individuals, was noted to have occurred in the recovery cages.

Figure 8 compares the three panel tests using the highest dose tested (0.10% a.i.) of permethrin WP and carbaryl WP. The general overall trends for the three tests appear to be the same for both compounds. The fastest breakdown of residual activity occurred on plywood panels (WA) against A. diaperinus adults. Test 1, where adults were exposed to treated styrofoam (SA), shows the longest overall residual activity. The residual activity in Test 3, where late instar larvae were exposed to treated styrofoam (SL), remains steady up until the fifth or sixth week, whereupon it begins to fluctuate wildly as the materials break down.

None of the materials or formulations had any adverse effect upon the surfaces to which they had been applied.

Topical Applications. The log dose-probit plot of the mortality data for permethrin is presented in Figure 9. The LD-50 for permethrin against A. diaperinus adults was shown to be 0.031 ug/beetle with 95% confidence intervals ranging from 0.016 ug/beetle to 0.061 ug/beetle. The LD-95 for

permethrin against A. diaperinus adults was shown to be 0.155 ug/beetle with 95% confidence intervals ranging from 0.075 ug/beetle to 0.974 ug/beetle.

The log dose-probit plot of the mortality data for tetrachlorvinphos is presented in Figure 10. The LD-50 for tetrachlorvinphos against A. diaperinus adults was shown to be 0.102 ug/beetle with 95% confidence intervals ranging from 0.088 ug/beetle to 0.117 ug/beetle. The LD-95 for tetrachlorvinphos against A. diaperinus adults was shown to be 0.222 ug/beetle with 95% confidence intervals ranging from 0.182 ug/beetle to 0.298 ug/beetle.

The log dose-probit plot of the mortality data for carbaryl is presented in Figure 11. The LD-50 for carbaryl against A. diaperinus adults was shown to be 0.556 ug/beetle with 95% confidence intervals ranging from 0.364 ug/beetle to 0.837 ug/beetle. The LD-95 for carbaryl against A. diaperinus adults was shown to be 3.136 ug/beetle with 95% confidence intervals ranging from 1.813 ug/beetle to 8.429 ug/beetle.

The toxic effects of each compound plus a control group were recorded. Immediately upon application of the acetone drop, beetles in the control group displayed an intial trauma and rolled over on their backs with legs paralyzed. The beetles recovered within 1-2 minutes. All other

treatments showed a similiar intial reaction to the application and this trauma most likely is attributable to the acetone carrier.

The onset of toxic symptoms was by far the most rapid with permethrin (0.1 ug/ul). Within 3-4 minutes post-application the insects began to show signs of intoxication. The beetles became still and their legs began to stiffen such that the body assumed an unnatural raised position. Within 5-10 minutes the beetles had fallen over onto their elytra accompanied with wild writhing and flailing of legs and mouthparts. Within minutes the movement in the prothoraic legs was slowed, being replaced with tremoring and slow deliberate movement of the front legs. The tremoring seemed to move posteriorally in a progressive fashion. At this time about half the beetles also began to regurgitate a clear fluid which quickly became viscous and amber in color, finally drying and encrusting the mouthparts such that they could no longer function. Within 30 minutes post-application, ovipositors and genitalia began to telescope in and out. Within 1 hour, general body movements had slowed and the extruded ovipositors had begun to collapse. Within two hours ovipositors had been retracted and very little movement was evident. At 24 h post-application all subjects were completely still.

Carbaryl (1.0 ug/ul) was second in the time of onset, with toxic symptoms beginning at ca. 30 minutes post-application. At this time there appeared to be slight paralysis of the limbs causing uncoordination. Within 45 minutes post-application movements became jerky and erratic, with beetles often walking backwards, slowing down or becoming still, then quickly running foward. Within 1 hour post-application there was 100% knockdown, with the beetles on their backs, legs and mouthparts moving agitatedly. There was no tremoring evident as in permethrin poisoning. At 5h post-application, two subjects had righted themselves and ran about hyperactively when probed. Those subjects on their backs displayed swollen abdomens, with the abdomenal sterna pulling away from the elytra exposing the membraneous pleuron and the terminal two or three abdomenal tergites. The scent glands were everted, however the ovipositors were not extruded. The abdomen of one subject was covered in a sticky fluid, presumably a benzoquinone compound discharged from the scent gland. At 24 h post-application six subjects (out of ten) had recovered and were moving about normally. Four subjects remained on their backs with legs occasionally moving slowly.

The onset of toxic symptoms for tetrachlorvinphos (0.3 ug/ul) did not occur until ca. 3h post-application.

Intoxication was similiar to that of carbaryl. Beetles appeared sluggish at first, moving their heads in an up and down fashion. Legs began to show paralysis and beetles displayed a loss of locomotory coordination, causing them to fall over on their backs. Once upon their backs, the insects moved their legs furiously but seemed unable to right themselves. As with carbaryl poisoning, no tremoring was observed, the abdomenal region began to swell as described, and everted scent glands were present but ovipositors did not become extruded. At 24 h post-application, two subjects had recovered, the other eight remaining on their backs only waving an occasional leg.

Discussion

Results of the panel tests indicate that both permethrin and carbaryl have considerably less residual activity against A. diaperinus when applied to plywood surfaces than when applied to polystyrene surfaces. Formulation may also be an important factor in these insecticides' practical application to various surfaces. The wettable powder formulation of permethrin resulted in excellent residual activity, but the emulsifiable concentrate formulation resulted in poor residual activity. Activity of both permethrin and carbaryl on styrofoam panels

appeared to differ depending upon the lifestage of A. diaperinus being tested. Adults were much more susceptible for a longer post-spray period than late instar larvae. This may have been due to the activity of the late instars burrowing into the treated styrofoam panels thus avoiding the toxic effects of the insecticide.

The stock colony used for Test 1 was gathered from a deep-pit layer house in Washington Co. near Abington, Va. The stock colony used for Test 2 and 3 was gathered from a deep-pit layer house in Amelia Co. near Jetersville, Va. It is possible that genetic differences between the two strains existed which produced differences in toxicity responses between adults and larvae on similiarly-treated panels. Different spray regimes within layer houses undoubtedly put different selective pressures on the mealworm populations occurring in different deep-pit houses. The operator of the Abington layer house reported spraying an average of every other week during the fly season, using mostly permethrin WP. The operator of the Jetersville layer house reported spraying up to 3-4 times a week during peak fly season, using a pyrethrum EC compound at low dosages (0.005%-0.01% a.i.). Because the insects used in Test 2 and 3 came from a population (Jetersville) that had been subjected to much heavier insecticide pressure than the insects in Test 1

(Abington population), the differences in toxicity response between adult and larvae presented in this study perhaps is an artifact from comparing resistant vs. non-resistant populations.

The insects used for the topical applications were all from the Jetersville population. The LD-50 values for permethrin (0.03 ug/beetle), tetrachlorvinphos (0.10 ug/beetle), and carbaryl (0.56mg/beetle) indicate that permethrin, being the most intrinsically toxic, may be the chemical of choice. Observations on the intoxication process of the materials show that recovery from toxic symptoms occurs with carbaryl even with 100% knockdown. No such recovery occurred with permethrin. The panel test data reveal that formulation problems, differences in surfaces, and differential mortality according to the lifestage of the insect may nullify the effectiveness of a material when applied in field situations. It is important to note that the regression slopes of both permethrin (1.03) and carbaryl (0.95) are much flatter than that of tetrachlorvinphos (2.11), implying that it would take a much larger increase in the number of molecules to get a small increase in toxicity with either permethrin or carbaryl than it would with tetrachlorvinphos. This points to an another consideration in the use of these compounds and that is the

possible development of resistance. Even though the log dose-probit line of permethrin lies to the left of the lines for tetrachlorvinphos and carbaryl, there seems to be less likelihood of resistance developing with the use of tetrachlorvinphos because its regression slope is over twice that of either permethrin or carbaryl. The response of A. diaperinus was much more homogenous to tetrachlorvinphos, suggesting that there are fewer individuals in the population that are resistant to tetrachlorvinphos, thus less chance of a resistant population developing to that compound. The panel tests indicate that resistance to both permethrin and carbaryl occur beween lesser mealworm populations gathered from different deep-pit layer houses known to implement different spray regimes. These results indicate that tetrachlorvinphos WP should be considered as the insecticide of choice to control A. diaperinus and to prevent the structural damage they cause in poultry houses.

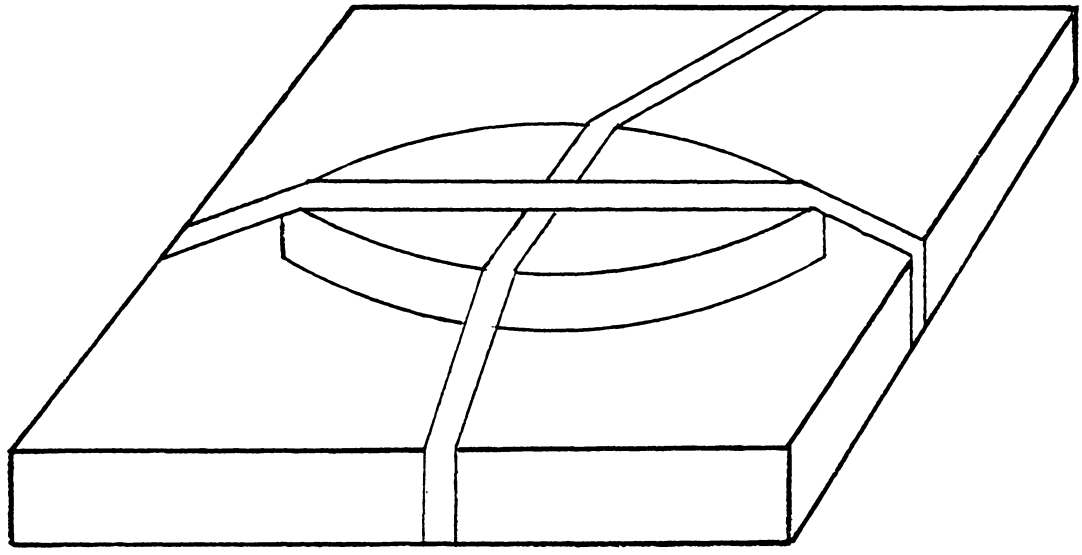

Figure 7. Experimental design for exposure tests to treated panels.

Table 9. Differences between 24hr and 48hr mortality counts for Alphitobius diaperinus adults when exposed for one hour to treated polystyrene panels.

DOSAGE AND FORMULATION	AVERAGE % MORTALITY 24hr	48hr	MEANS STATISTICALLY DIFFERENT[1]
0.50% tetrachlorvinphos WP	100.00	99.55	NO
0.10% permethrin WP	99.58	96.82	NO
0.05% permethrin WP	97.80	85.53	NO
0.01% permethrin WP	32.80	12.22	YES
0.10% permethrin EC	50.95	18.18	YES
0.05% permethrin EC	39.25	10.58	YES
0.01% permethrin EC	18.17	11.02	NO
0.10% carbaryl WP	96.54	92.06	NO
0.05% carbaryl WP	75.80	69.63	NO
0.01% carbaryl WP	25.48	21.32	NO

[1]alpha=0.05. Percent mortality represents the average corrected mortalities for the entire 21-week test period.

Table 10. Average corrected percent mortality of Alphitobius diaperinus adults exposed for one hour to treated polystyrene panels.

	MEAN % MORTALITIES									
WEEK	tet W 0.50	per W 0.10	per E 0.10	car W 0.10	per W 0.05	per E 0.05	car W 0.05	per W 0.01	per E 0.01	car W 0.01
0	100.0a	100.0a	77.6ab	100.0a	96.3a	40.2bc	100.0a	6.9c	40.5bc	35.3c
+1	100.0a	100.0a	0.0c	96.4a	88.9a	0.0c	89.3a	0.0c	0.0c	20.2b
+2	100.0a	100.0a	10.7c	100.0a	100.0a	1.2c	96.7a	14.3c	2.4c	30.0b
+4	100.0a	96.7a	0.0b	90.0a	80.0a	6.7b	86.7a	13.3b	0.0b	23.3b
+6	100.0a	96.3a	0.0b	80.7a	85.2a	0.0b	22.9b	25.9b	7.4b	19.1b
+8	100.0a	91.4a	2.3c	-	93.2a	0.0c	-	27.6b	0.0c	-
+11	-	-	-	85.2a	-	-	22.2b	-	-	0.0b
+17	100.0a	96.1a	2.0c	-	58.4b	2.0c	-	0.0c	15.7c	-
+21	100.0a	94.2a	52.9bc	-	82.3ab	34.6cd	-	9.8d	22.2cd	-

[1]means within rows with common letter are not significantly different at the 0.05 level. Treatments are: tetrachlorvinphos WP 0.50% a.i. (tetW 0.50), permethrin WP 0.10% a.i. (perW 0.10), 0.05% a.i. (perW 0.05), 0.01% a.i. (perW 0.01), permethrin EC 0.10% a.i. (perE 0.10), 0.05% a.i. (perE 0.05), 0.01% a.i. (perE 0.01), and carbaryl WP 0.10% a.i. (carW 0.10), 0.05% a.i. (carW 0.05), 0.01% a.i. (carW 0.01).

Table 11. Average corrected percent mortality of Alphitobius diaperinus adults exposed for one hour to treated plywood panels.

WEEK	MEAN % MORTALITIES						
	tetW 0.50	perW 0.10	carW 0.10	perW 0.05	carW 0.05	perW 0.01	carW 0.01
0	100.0a	87.0ab	82.8ab	75.2b	92.3ab	22.7c	2.0c
+1	100.0a	93.6a	100.0a	72.0b	89.5a	27.0c	8.7d
+2	100.0a	81.7a	93.3a	56.7b	58.3b	11.7c	6.7c
+3	100.0a	77.2b	88.3ab	45.0c	43.3c	15.0d	3.5d
+5	100.0a	57.4bc	77.4ab	46.3cd	68.7bc	23.5de	3.5e
+6	100.0a	47.4b	40.5b	14.7d	26.9c	0.0e	0.0e
+11	100.0a	13.0b	3.7c	3.7c	0.0c	0.0c	3.7c

[1]means within rows with common letter are not significantly different at the 0.05 level. Treatments are: tetrachlorvinphos 0.5% a.i. (tet 0.50), permethrin 0.10% a.i. (per 0.10), 0.05% a.i. (per 0.05), 0.01% a.i. (per 0.01), and carbaryl 0.10% a.i. (car 0.10), 0.05% a.i. (car 0.05), 0.01% a.i. (car 0.01).

Table 12. Average corrected percent mortality of Alphitobius diaperinus late instar larvae exposed for one hour to treated polystyrene panels.

WEEK	MEAN % MORTALITIES tetW 0.50	perW 0.10	carW 0.10	perW 0.05	carW 0.05	perW 0.01	carW 0.01
0	100.0a	100.0a	100.0a	100.0a	96.4ab	59.4c	81.9b
+1	100.0a	100.0a	98.3a	100.0a	95.0a	70.0b	23.3c
+2	100.0a	100.0a	100.0a	100.0a	100.0a	100.0a	66.7b
+3	100.0a	100.0a	100.0a	100.0a	100.0a	80.0b	46.7b
+5	100.0a	100.0a	100.0a	92.6a	85.7a	13.5b	11.2b
+6	100.0a	53.3b	66.7b	40.0b	60.0b	6.7c	0.0c
+7	100.0a	100.0a	100.0a	56.7b	36.7b	3.3c	0.0c
+8	100.0a	73.3b	40.0c	31.7c	7.5d	0.0d	0.0d
+9	100.0a	46.7b	6.7d	16.7c	0.0d	3.3d	6.7d
+10	100.0a	66.7bc	86.7ab	60.0c	33.3d	16.7d	10.0d
+11	96.7a	100.0a	54.2b	20.0c	13.3cd	6.7cd	3.3d
+12	93.3a	56.7b	6.7d	30.0c	0.0d	0.0d	0.0d
+16	86.7a	83.3a	53.3b	43.3b	0.0c	10.0c	0.0c
+17	100.0a	100.0a	34.5b	30.7b	0.0c	23.8b	0.0c
+18	90.0a	20.0b	10.0bc	0.0c	0.0c	0.0c	0.0c
+19	100.0a	30.0b	13.3c	0.0d	10.0cd	10.0cd	0.0d

[1] means within rows with common letter are not significantly different at the 0.05 level. Treatments are: tetrachlorvinphos 0.50% a.i. (tet 0.50), permethrin 0.10% a.i. (per 0.10), 0.05% a.i. (per 0.05), 0.01% a.i. (per 0.01), and carbaryl 0.10% a.i. (car 0.10), 0.05% a.i. (car 0.05), 0.01% a.i. (car 0.01).

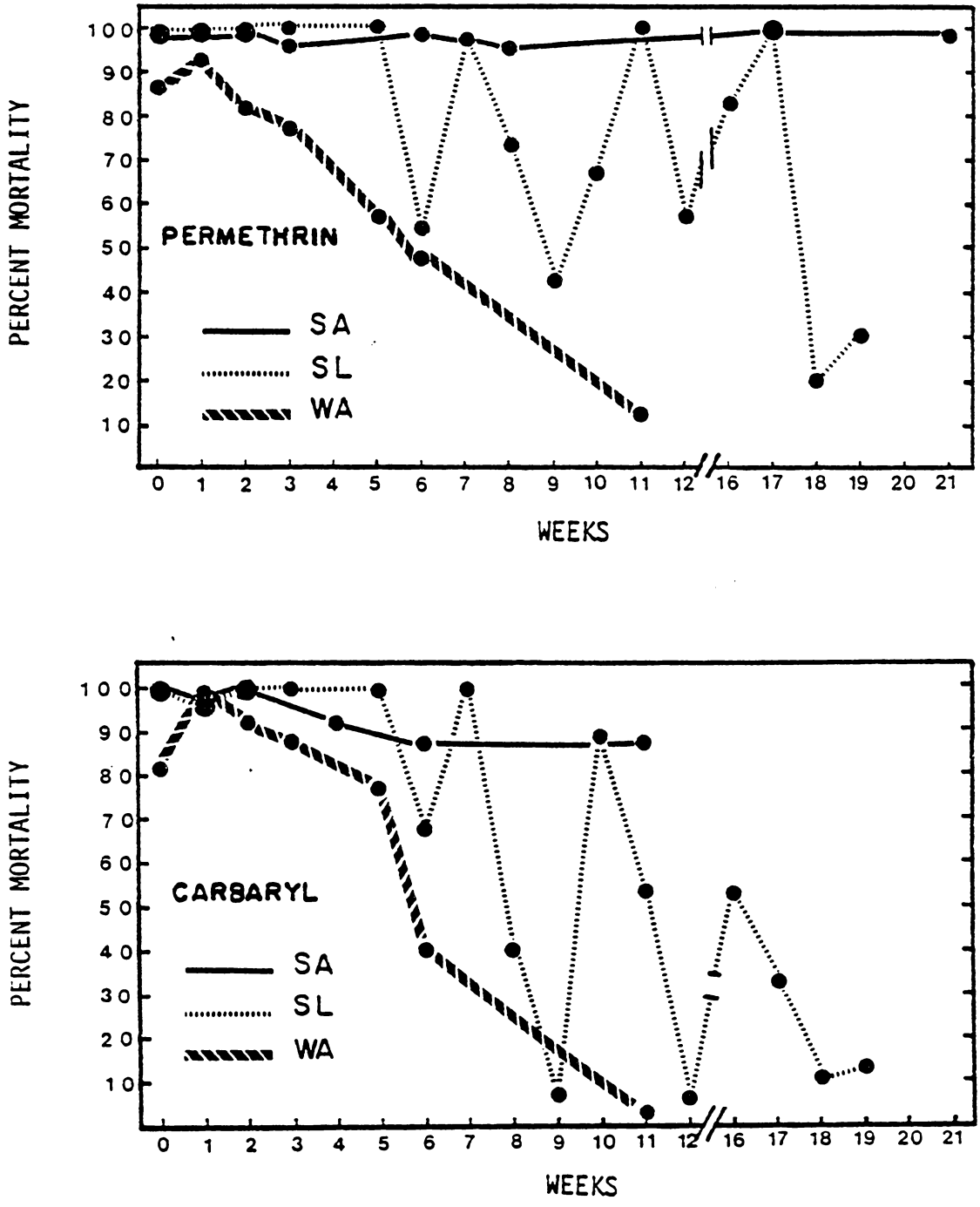

Figure 8. Mean corrected percent mortalities of Alphitobius diaperinus exposed for one hour to surfaces treated with 2ml per ft^2 of permethrin WP 0.1% a.i. and carbaryl WP 0.1% a.i. SA=adult beetles on treated styrofoam. SL=late instar larvae on treated styrofoam. WA=adult beetles on treated plywood.

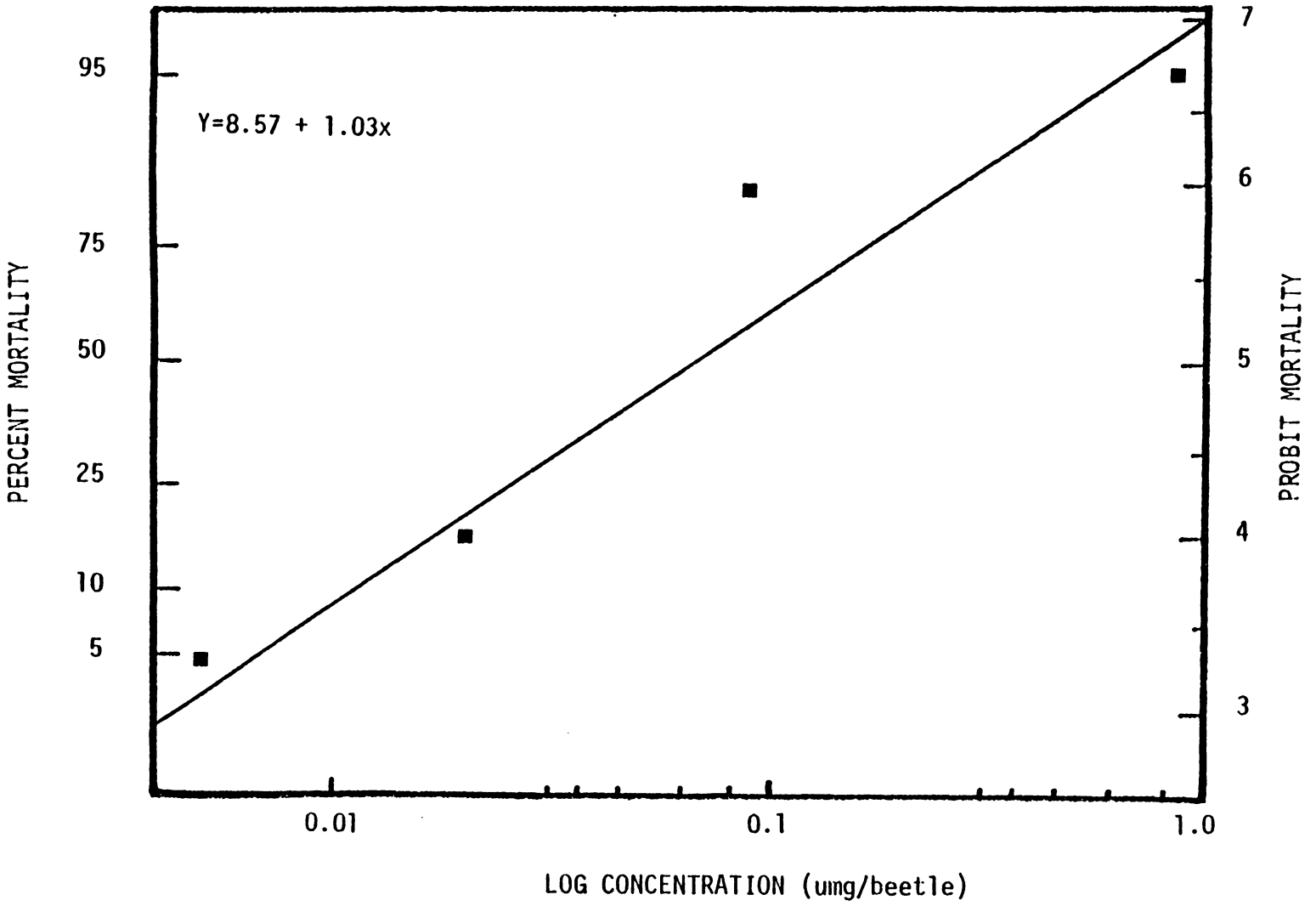

Figure 9. Response of Alphitobius diaperinus (Panzer) to topical application of permethrin.

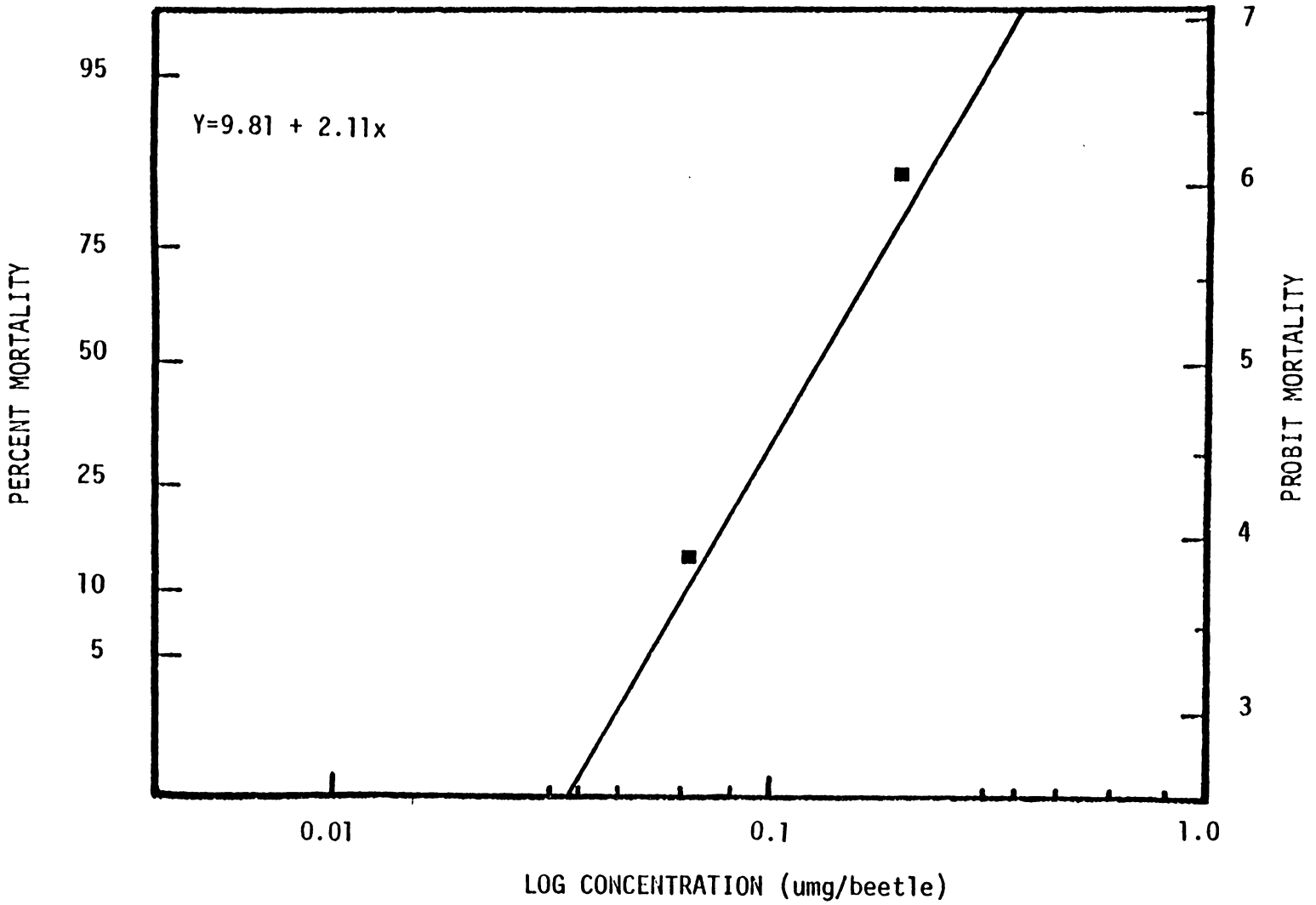

Figure 10 Response of Alphitobius diaperinus (Panzer) to topical application of tetrachlorvinphos.

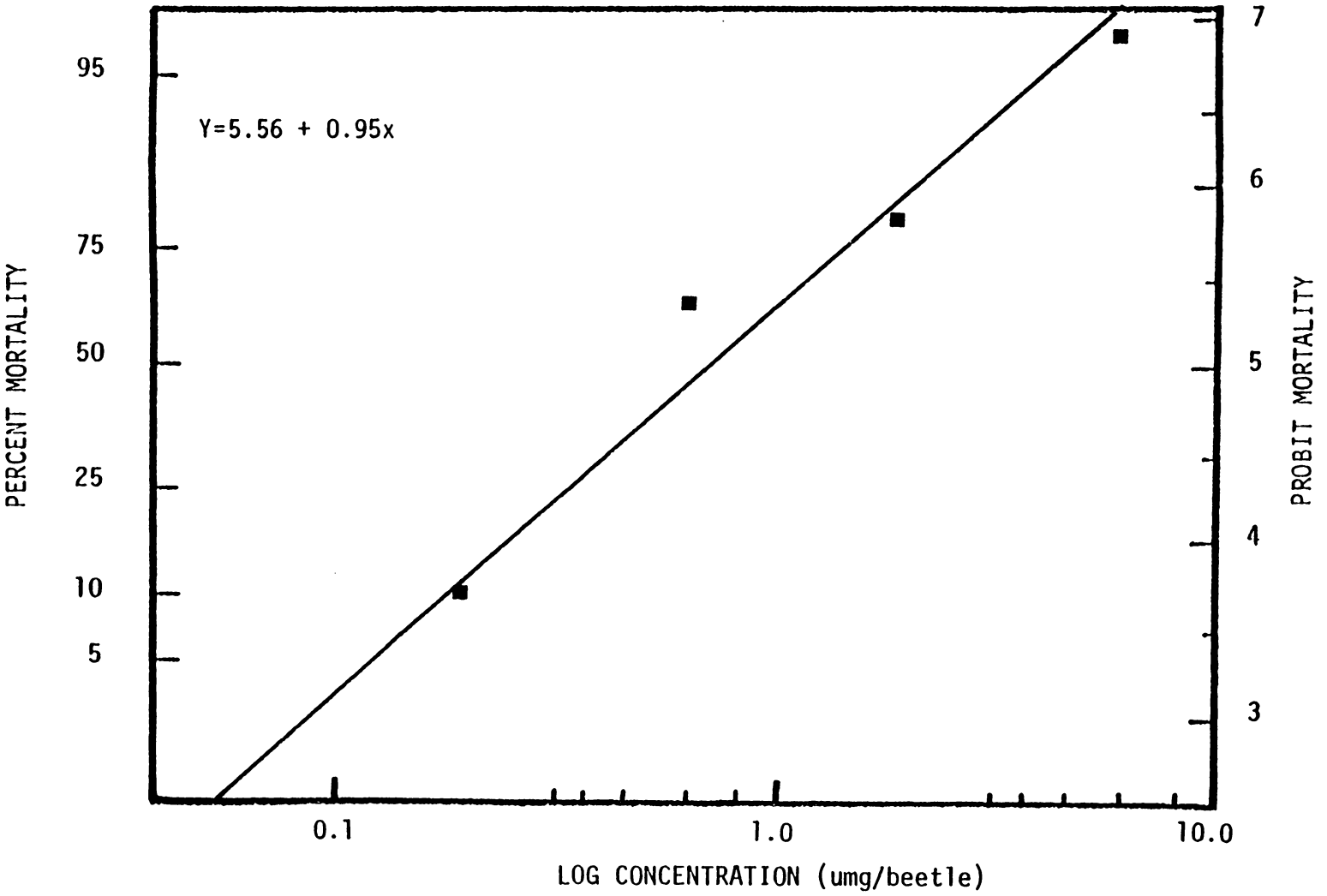

Figure 11. Response of Alphitobius diaperinus (Panzer) to topical application of carbaryl.

VI. SUMMARY

The lesser mealworm, Alphitobius diaperinus, causes structural damage in deep-pit layer houses by tunneling into exposed panels of polystyrene and polyurethane insulation. Research was conducted to; 1) follow the chronological succession of lifestages that occurs in polystyrene, polyurethane, and fiberglass insulation, 2) observe behavior with emphasis on cannibalism and house fly predation, and 3) conduct toxicity studies with three insecticides (permethrin, carbaryl, and tetrachlorvinphos) approved for use in Va. poultry houses.

A distinct pattern of infestation occurred in polystyrene and polyurethane. Late instar larvae intiated tunneling behavior in order to pupate, and intial adult activity was restricted to oviposition upon the panel surface. Although many eggs hatched, the early instar larvae did not stay on the panels. Middle instar larvae were not present. After ca. 13 days, adults began to move into the tunnels, depreciating the insulation's suitability as pupation sites. Subsequently, the numbers of late instar larvae, pupae, and eggs declined. All lifestages invaded exposed fiberglass and it was not used by late instar larvae as pupation sites.

All lifestages of A. diaperinus were found to be cannibalistic on pupae. Cannibalism, particularly at high population densities, may play an important role in causing late instar larvae, seeking protected pupation sites, to tunnel into polystyrene or polyurethane insulation. Although preliminary tests show that adult mealworms will consume third instar house fly larvae and prepupae, no significant predation of the house fly by A. diaperinus could be detected under simulated natural conditions. Flight and mating activity occurred with A. diaperinus in early summer (May-June). It is suggested that adult flight, from natural reservoirs or other poultry houses, may be one way in which A. diaperinus colonizes newly-built poultry houses.

Tetrachlorvinphos showed longer residual effectiveness against A. diaperinus as a surface spray on polystyrene (19wk) and plywood (11wk) than did permethrin or carbaryl. Wettable powder formulations of permethrin and carbaryl, when applied to polystyrene, were comparable in residual activity. Activity of both compounds was less when applied to unpainted plywood. The emulsifiable concentrate formulation of permethrin at dosages up to 0.10% a.i. proved to be ineffective. The slope of the regression equation for tetrachlorvinphos (2.11) obtained from topical application

studies reveals there is less chance of the lesser mealworm developing resistance to it than with either permethrin (1.03) or carbaryl (0.95).

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VIII. APPENDICES

APPENDIX I

REPORTED DISTRIBUTION OF *ALPHITOBIUS DIAPERINUS* IN LITERATURE

Association Code:
S = *A. diaperinus* associated with stored products.
P = *A. diaperinus* associated with poultry production.
N = *A. diaperinus* associated with natural habitats, wild birds and mammals.

REGION & COUNTRY	ASSOCIATION	CITATION
NEARCTIC ZOOGEOGRAPHICAL REGION		
Canada	S:	Sinha 1965
Continental U.S.A.	S:	Back and Cotton 1962, Cotton and Good 1937, Douglas 1941, Ebeling 1975, Gould 1948, Keifer 1935, Rilett and Weigel 1956, Strong 1970.
	P:	Bray et al. 1968, Case and Ackert 1940, Collison 1980, De las Casas et al. 1968, 1969, 1970, 1972a, 1972b, 1973, 1974, 1976, Dunning et al. 1978, Eidson et al. 1965 1966, Eugenio et al. 1970, Gall 1980, Green, M. 1980, Gould 1948, Harding and Bissell 1958, Harein et al. 1969, 1970, 1972, Harris 1966, Lancaster et al. 1967, 1969, Legner et al. 1975, Legner and Olton 1968, McCreary and Catts 1954, Pfieffer 1954, Pfieffer and Axtell 1980, Preiss 1969, Schmittle 1966, Silberman and Schmittle 1967, Simco et al. 1966, Smith 1981, Snedeker et al. 1967, Spilman 1968, Thornberry 1978.
	N:	Davis et al. 1962, Levi 1957, Linsely 1944, Thompson 1944.

REPORTED DISTRIBUTION OF *ALPHITOBIUS DIAPERINUS* IN LITERATURE

REGION & COUNTRY	ASSOCIATION	CITATION
NEOTROPIC ZOOGEOGRAPHICAL REGION		
Argentina	S:	Green, M. 1980.
Barbados	S:	Bovell 1917.
Chile	P:	Pena 1973.
Cuba	P:	Blahutiak and Barus 1970.
Jamaica	S:	McFarlane 1963.
Puerto Rico	P:	Legner and Olton 1970.
PALEARCTIC ZOOGEOGRAPHICAL REGION		
Britain	S:	Freeman 1964, Joy 1932, Soloman and Adamson 1955.
	P:	Green, D. B. 1980, Green, M. 1980, Halstead 1975, Jerrard and Wildey 1980.
Denmark	P:	Green, M. 1980, Halstead 1975.
Finland	P:	Silfverberg 1979.
France	S:	Lepesme 1944.
	P:	Chaix 1980, Torc'h 1979.
Germany	S:	Piltz 1960, Tischler 1937, Weiss 1973, Zacher 1938.
	P:	Geissler and Kosters 1972, Gersdorf 1969, 1970, Heimbucher and Kutzer 1979, Hilbrich 1978, Lohren 1972, Weiss 1973.
Hungary	P:	Nemeseri and Gestessy 1973, Swatonek 1970.
Ireland	P:	O'Farrell and Butler 1948, O'Mahoney 1950.
Netherlands	P:	Bouvy 1973.
U.S.S.R.	P:	Koszlov 1970.
MEDITERRANEAN ZOOGEOGRAPHICAL REGION		
Egypt	S:	Salama and Salem 1973.
	N:	Guirgis 1971.
Spain	S:	Espanol 1956.

REPORTED DISTRIBUTION OF *ALPHITOBIUS DIAPERINUS* IN LITERATURE

REGION & COUNTRY	ASSOCIATION	CITATION
ETHIOPIAN ZOOGEOGRAPHICAL REGION		
Israel	P:	Legner and Olton 1970.
Kenya	N:	McFarlane 1971.
Mozambique	N:	Scott 1928.
Nigeria	S:	Halliday and Kazaure 1969.
Senegal	S:	Roubaud 1916.
Sudan	P:	Elowni and Elbihari 1979.
	N:	Buck 1956, Johnston 1933, Lewis 1958.
Tanzania	N:	Britton 1940.
AUSTRALASIAN ZOOGEOGRAPHICAL REGION		
American Samoa	P:	Legner and Olton 1970.
Australia	S:	McCarthy 1922.
Hawaii	P:	Alicata 1947, Cuckler and Alicata 1944.
New Zealand	P:	Dale et al. 1976.
ORIENTAL ZOOGEOGRAPHICAL REGION		
Borneo	S:	Aitken 1975.
Burma	S:	Aitken 1975.
India	S:	Aitken 1975, Green, M. 1980, Pajni and Gill 1974, Sarin and Saxena 1975.
Japan	S:	Nakao 1975, Yoshida 1975.
	P:	Ichinose et al. 1980.
Malaya	S:	Aitken 1975.
Phillippines	N:	Crook et al. 1981.
Sri Lanka	S:	Rajendra 1979.
Taiwan	S:	Li 1953.
Thailand	S:	Aitken 1975.

APPENDIX II

STORED PRODUCTS COMMODITIES FROM WHICH
ALPHITOBIUS DIAPERINUS HAS BEEN REPORTED TO INFEST

Commodity	Citations
animal feeds	Aitken 1975, Fowler 1891, McFarlane 1963.
animal products (hides, bones, etc.)	Aitken 1975, Espanol 1956, Green, M. 1980.
barley	Sarin and Saxena 1975.
bread (dried hard bread, moldy)	Tischler 1937.
cereal and cereal products (damp or moldy)	Aitken 1975, Back and Cotton 1962, Ebeling 1925, Espanol 1956.
cocoa beans	Aitken 1975.
copra	Aitken 1975.
cotton seed (decaying)	Bovell 1917.
cowpea	Sarin and Saxena 1975.
flour (damp or moldy)	Back and Cotton 1962, Cotton and Good 1932, Espanol 1956, Fowler 1891, Joy 1932, Salama and Salem 1973, Tischler 1937.
grain and grain by-products (damp or moldy)	Aitken 1975, Back and Cotton 1962, Cotton and Good 1937, Green, M. 1980, Gould 1948 (corn), Halstead 1975, McCarthy 1922, Plitz 1960, Sarin and Saxena 1975 (wheat).
ground nuts (peanuts)	Halliday and Kazaure 1969 (cited *A. laevigatus*), Roubaud 1916.

STORED PRODUCT COMMODITIES FROM WHICH
ALPHHITOBIUS DIAPERINUS HAS BEEN REPORTED TO INFEST

Commodity	Citation
illipenuts (family Sapotaceae)	Aitken 1975.
milk powder	Nakao 1975, Yoshida 1975.
oilcake	Aitken 1975, Green, M. 1980.
oilseeds	Aitken 1975, Green, M. 1980.
palm fruits	Lepesme 1947.
pimento	McFarlane 1963.
rice and rice by-products	Aitken 1975, Douglas 1941, Li 1953, McFarlane 1963, Sarin and Saxena 1975.
sugarcane (germinating shoots)	Rajendra 1979.
sunflower seeds	Atanasov 1974.
urd (black chickpea)	Sarin and Saxena 1975.

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BIOLOGY AND CONTROL OF THE LESSER MEALWORM: *ALPHITOBIUS DIAPERINUS* A STRUCTURAL PEST IN POULTRY HOUSES

by

Jefferson Archer Vaughan

(ABSTRACT)

Late instar larval *Alphitobius diaperinus* intiated tunneling damage to polystyrene insulation in search of protected pupation sites. Adult females oviposited upon the surface of the styrofoam but early instar larvae left the styrofoam soon after hatching. Within twenty days, large numbers of adults moved into the styrofoam, expanding the tunnels and depreciating the panels' suitability as pupation sites for the late instar larvae. No oviposition occurred within the tunnels.

All lifestages of *Alphitobius diaperinus* were found to be cannibalistic. Although preliminary tests show that adult mealworms will consume house fly late instar larvae and prepupae, no significant predation of the house fly by *A. diaperinus* could be detected under simulated natural conditions at the population density tested.

Wettable powder formulations of permethrin and carbaryl when sprayed upon styrofoam were comparable in residual activity. Residual activity of both compounds was less when

applied to unpainted plywood. The emulsifiable concentrate formulation of permethrin proved to be ineffective. Regression slopes from topical application studies reveal that tetrachlorvinphos may be the material of choice against A. diaperinus because there is less chance of the lesser mealworm developing resistance to tetrachlorvinphos than with either permethrin or carbaryl. Tetrachlorvinphos also showed longer residual effectiveness against A. diaperinus as a surface spray on styrofoam and plywood than did permethrin or carbaryl.