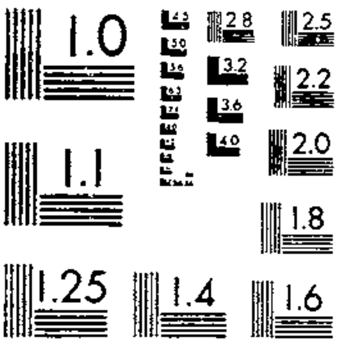
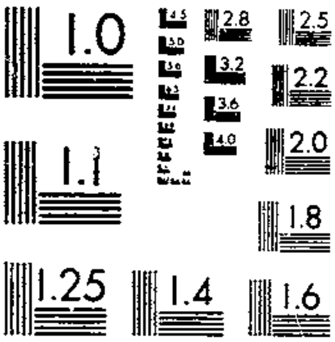
TB 336 (1932) USDA TECHNICAL BULLETINS UPDATA
ATTRACTANTS FOR THE FLYING GIPSY MOTHS AS AN AID IN LOCATING NEW
COLLINS, C.W. ; POTTS, S.F. 1 OF 1

START



MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A



MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

TECHNICAL BULLETIN No. 336 — DECEMBER, 1932

UNITED STATES DEPARTMENT OF AGRICULTURE
WASHINGTON, D. C.

ATTRACTANTS FOR THE FLYING GIPSY MOTHS AS AN AID IN LOCATING NEW INFESTATIONS

By C. W. COLLINS, *Senior Entomologist*, and S. F. POTTS, *Assistant Entomologist, Division of Forest Insects, Bureau of Entomology*[1]

CONTENTS

INTRODUCTION

Experiments with attractants for the male of the gipsy moth, *Porthetria dispar* L., were begun with the hope of reducing infestations by attracting the males in a given area to traps containing caged females, thus leaving the females in the area unfertilized. Though this idea seemed plausible, the plan was not successful, and the work was directed toward the following objectives: (1) To locate new infestations as an aid to scouting and eradication work, (2) to study the flight and habits of the male moths, (3) to substitute extracts of female "tips"[2] for living females in the traps, thus elimi-

[1] The writers are indebted to A. F. Burgess for general direction and suggestions while he was connected with the Bureau of Entomology, and for continued cooperation since his connection with the Bureau of Plant Quarantine; and to H. L. Blaisdell, S. S. Crossman, and H. A. Ames, of the Bureau of Plant Quarantine, for their hearty cooperation and assistance. L. H. Worthley and H. L. McIntyre, formerly of the Bureau of Entomology, assisted in the execution of some of the large field experiments. Several workers at the gipsy-moth laboratory, Melrose Highlands, Mass., gave valuable assistance, especially C. E. Hood, J. E. R. Holbrook, R. Wooldridge, P. B. Dowden, D. F. Barnes, R. L. Wallis, R. R. Whitten, and F. W. Graham (formerly of the Bureau of Entomology). E. P. Kohler, professor of organic chemistry at Harvard University, rendered valuable cooperation in connection with the chemical studies, as did also three other chemists mentioned in connection with their reports.

[2] "Tip" refers to the posterior end of the abdomen comprising the last segments and genitalia.

nating the danger of starting new infestations in a possibly free area, (4) to locate the organ or glands which produce the attracting substance, (5) to determine the possibility of isolating, studying, and producing the attractive substance in quantity synthetically and cheaply, and (6) to determine the value of this information for work on other insect species.

Though considerable has been written on the sense of smell in mammals, this subject has been much less thoroughly investigated than have the senses of sight, hearing, and touch. Taste, which is associated with smell, has likewise received relatively little attention. The sense of smell is continually employed throughout most of the animal kingdom, in connection with obtaining food, protection, recognition, communication, and mating. Studies of the sense of smell in insects have, for the most part, been centered on recognition, as among the bees and ants, or in connection with taste as related to obtaining food. It has been known for centuries that male moths may be attracted from a distance by females of the same species, but this fact has been mainly taken for granted and has been given very little detailed study.

The female gipsy moth does not fly, but the male is a strong flier and is able to locate the female by means of scent. The scent is given off from the vicinity of the vaginal opening of the female and is picked up by the male through its antennae. The male proceeds to find the female by means of zigzag flight, usually against the wind carrying the scent. According to Forbush and Fernald [3] the male antennae are very strongly bipectinated, the pectinations being somewhat curved, and tapering slightly toward the outer end, where they terminate in a tooth on one side, and a much longer spine on the other. In the female the pectinations are very much shorter and stouter.

To ascertain whether any advantage could be taken of these habits by attracting the males to traps containing unfertilized females, A. H. Kirkland began some experiments in eastern Massachusetts in 1893,[4] which were continued by H. N. Reid, to determine whether it were possible to trap enough males in a heavy infestation to reduce the fertilization of eggs enough so that a marked reduction in the number of moths and the degree of infestation could be expected the following season. These experiments were carefully planned and executed, but as a final result only 2.4 per cent of the egg clusters deposited in the area where the traps were exposed proved to be infertile. Since some infertile eggs are generally found in gipsy-moth infestations, this percentage has little significance.

Up to 1913 the area infested by the gipsy moth had continued to increase, and isolated colonies were sometimes found in unexpected places well beyond the supposed border of infestation. Such colonies were difficult and expensive to locate by scouting without clues as to their location or existence. A. F. Burgess, at that time in charge of the gipsy-moth investigations for the Bureau of Entomol-

---

[3] Forbush, E. H., and Fernald, C. H., THE GYPSY MOTH. (PORTHETRIA DISPAR LINN.) A REPORT OF THE WORK OF DESTROYING THE INSECT IN THE COMMONWEALTH OF MASSACHUSETTS TOGETHER WITH AN ACCOUNT OF ITS HISTORY AND HABITS BOTH IN MASSACHUSETTS AND IN EUROPE. P. 339-340. Boston. 1896.

[4] Forbush, E. H., and Fernald, C. H. Op. cit., p. 345-363.

ogy, decided that some experiments should be conducted to facilitate finding these colonies, and the senior writer took up the investigations at that time.

TYPES OF CAGES AND MARKING AND LIBERATION OF MALES

The first cage used in 1913 was made from a wooden box 15 by 4 by 4 inches. The top and bottom were replaced with a double wall of 20-mesh wire screening with one-half inch space between walls to prevent mating by the inclosed females with males that might be attracted to the cages. Later, when a large number of these cages were required for use in border territory infested by the gipsy moth, it was necessary to reduce the size, and accordingly a cage 5 by 3½ inches made of ½-inch wood stock, with a clearance space between top and bottom of 1¾ inches, was found advantageous. A covered circular hole 1½ inches in diameter in the top served for the introduction of female pupae, or moths. A wooden strip of ½-inch stock served to hold the top and bottom together and to attach the cage to a tree. Double walls one-half inch apart made of fly wire screen composed the four sides. This insured good circulation of air to carry the scent and yet prevented mating with outside males. (Fig. 1.)

FIGURE 1.—Type of cage used in early experiments and as check cages to hold living females of the gipsy moth. The illustration shows the trap number and the males caught in the sticky material in a single day

After it was found that males could be attracted by means of an extract from the tips of abdomens of females, a 3-ounce tin salve box (fig. 2) was used as a container. From four to six small holes, in pairs or groups of three, were drilled in each side of the tin for the passage of air to evaporate the solvent and attractive substance. During heavy rains, more or less water entered through these holes, and this necessitated a change of method. After this experience a 3-ounce uncovered can filled with cotton (fig. 3) was used. The extract was poured on the cotton and the can then inverted and fastened to the tree in this position. The inverted open can protected the extract from rains, and allowed the odors to escape more evenly. A sticky substance applied to the tree trunk around the trap served to catch the males coming to and hovering about the trap.

The cages or traps were usually placed near the roadside in the edges of woodland, neglected orchards, or sprout growth, and along the edges of fields, because such localities were more apt to be infested and could also be more easily visited. The cage was fastened on the side of a tree away from the road so as not to attract attention. During seasons when a large number of cages were put out they were placed along roadsides three-fourths mile or 1 mile apart, and in special experiments and locations as near together as one-fourth to one-half mile.

FIGURE 2.—A covered tin salve box as used in early experiments with extracts. A single day's catch of males is shown in the sticky material

In obtaining data on distances or direction males were attracted (or flew to find females) it was necessary to mark and liberate many males, and distinctive markings were necessary to indicate the various liberation points. Aniline dyes mixed with 70 per cent alcohol were used in 1913 for this purpose. The different colors were applied to certain wings by means of a small camel's-hair brush. The males flutter about considerably in the boxes, and marks made with the above mixture would sometimes spread to other wings than those marked or to parts of the body before drying, making its use unsatisfactory. Artists' oil paints thinned with gasoline were later used in the same manner and with much better results. The colors did not spread so rapidly and dried much more quickly.

In 1928 male pupae were put in a darkened box with a cone at one end, through which the males, soon after issuing, crawled toward the light. A wick extending from a bottle of dye solution was held in the cone, and as the males crawled over the wick in order to get through the cone they became marked with the dye. When they were caught at the trap a solvent was used to dissolve the sticky substance and dye in order to determine the source of the moths. The marked males were liberated at various points from one-sixteenth mile to 1 mile distant from the central female cage, and notes were taken on the time of flight and direction and distance from which they were attracted to the trap.

LIVING FEMALE MOTHS AS SOURCE OF ATTRACTION

During July and August, 1913, some preliminary experiments were conducted near the western border of the New England area then known to be infested.[5] The purpose of these experiments was

FIGURE 3.—An open can filled with cotton into which an attractive solution has been poured: A, In position for receiving the attractant; B, inverted. Adhering to the sticky material is shown a day's catch of males.

to obtain data on the number of males that might be attracted to a given point, the idea being to follow up the catches by scouting to determine the distance to the nearest infestation and the direction from which the males came. Ten cages were put out in as many locations, each location being a few miles from any known infestation. Twenty female pupae, from which 10 to 13 female moths issued, were inclosed in each cage. From 1 to 6 males were caught at six of the

[5] In 1913 this area in Massachusetts extended from the northeast corner of Connecticut in a zigzag line to Northfield, Mass., where the Connecticut River enters Massachusetts, thence northeasterly in New Hampshire to Ellsworth and Thornton in Grafton County, thence eastward into Maine.

cages. Detailed scouting was later done around two of these six cage sites to a distance of one-half mile and 2 miles, respectively, but no infestations were found.

Some similar experiments were conducted in 1914. Ninety-three cages were distributed in 23 towns of Maine, New Hampshire, and Massachusetts about the middle of July and removed the middle of August. One or two males were caught at 25 of these cages. Some intensive scouting was done the following fall in a few selected locations to determine the nearest infestations. A rather interesting find was made in Rumney, N. H., consisting of four infestations, located as follows with reference to cages: One infertile egg cluster two-thirds of a mile south of a cage which attracted 2 males; one fertile egg cluster 1½ miles east of a cage which attracted 1 male; another fertile egg cluster two-thirds of a mile southeast as well as a trace of an egg cluster 1⅓ miles southwest of a cage which attracted 2 males. The first two infestations were found by the regular scouting crews examining favorable tree growth along roadsides, edges of woodlands, and all scattered trees in the open, and the last two by a special scouting crew that carefully examined the whole of the wooded area.

One infestation in Westmoreland, N. H., was 1⅛ miles from a cage that attracted males, but the infestation was not located until the second year of scouting after the catch was made. Other interesting catches of males were made in Stoddard and in Gilsum, N. H., in 1914, and near-by infestations were found during the scouting seasons of 1914–15 and 1915–16. Infestations were located in one of those years within 500 yards, one-half mile, and 1 mile of the attracting cages.

Three males were attracted to a cage near the town line of Newport and Croyden, N. H., in 1913. Subsequent scouting did not reveal any infestations that season. Another cage was placed in the same location in 1914 and a circle of cages around it as a center, ranging from three-fourths mile to 1½ miles distant. Owing to exceedingly poor issuance of females from the pupae, poor results were secured. One cage, however, attracted 1 male, and subsequently one fertile egg cluster was found one-fourth mile south. To show that the attraction of 3 males to the first cage in 1913 was indicative of general conditions, an infestation of 66 fertile egg clusters was found about 2 miles east during the 1914–15 scouting, and two egg clusters at one-half and three-fourths mile distant. Several additional egg clusters were found distributed over an area within 1½ miles of the 1913 cage site.[6]

FLIGHT OF MALES AND PROPORTION FINDING FEMALES

Tests to obtain data on the flight of males and the proportion finding females from given distances were conducted at North Dartmouth, Mass., in August, 1913. In these tests fresh males were marked with aniline dyes in 70 per cent alcohol. The marking was done in the early forenoon, when the males were least active, with different colors on the right or left fore wing to denote liberation

[6] Though no intensive scouting was done around cages which did not attract males it is probable that some infestations existed within the same distances from those not attracting as from those that did. Later experiments have shown that these variable results may arise from such conditions as unfavorable topography, wind, temperature, or dense tree growth.

point. In all, 275 males were liberated three-fourths mile and 1 mile in different directions from a central cage containing 35 virgin females.

One unmarked male was taken at the central cage, and after scouting a small natural infestation was found one-fourth mile east from which it may have come. Two days later a marked male from the liberation three-fourths mile north was taken. The counter attraction offered by such natural infestations may account for the fact that no other of the marked males were caught.

On July 9, 1914, a cage containing 20 female gipsy-moth pupae was attached to a post on Lunging Island, Isles of Shoals, off the coast of New Hampshire. This island, which is small and rocky, was not infested, the nearest infestations being on White Island, one-half mile distant; Appledore Island, five-eighths mile distant; and Smutty Nose Island, three-fourths mile distant. On August 8, 33 other females were added to the cage. On August 31 the cage was examined, and 79 males had been caught. It was interesting that all the males were caught on the side of the trap toward the infested islands.

A similar test was conducted at Race Point, Provincetown, Mass., August 9 to 25, 1915. A cage containing 20 females was attached to a telephone pole on the beach near the lighthouse. The nearest infestations were in woodlands beyond some sand dunes from 1¼ to 1½ miles to the east and southeast and from 1¾ to 2 miles to the northeast. Nine males were caught this year, and in a repetition of this experiment in August, 1917, 59 males were caught.

Experiments to obtain more data were conducted on Lynn Marshes near Lynn, Mass., in 1915. This is a broad expanse of treeless, salt marshland relatively free from gipsy-moth infestation. Six experiments were conducted from August 10 to 15 by liberating marked males with, against, and at right angles to the winds at distances from 150 yards to three-fourths mile. Results from liberations at 150 yards and one-fourth mile distant showed that the males fly against the wind in almost all cases. Of 362 marked males, 27, or one-thirteenth of the number liberated, were caught. Ninety-nine unmarked males, however, came to cages from near-by infestations 150 yards to 1 mile distant. Four of 18 marked males liberated one-fourth mile to the northwest were recovered between 45 minutes and 3 hours and 40 minutes after liberation. The marked males all came flying against the wind.

Other experiments were also conducted on marshlands at the same time by liberating 72 marked males about noon leeward of the wind, and three-fourths mile distant from a cage of 40 virgin females. The cage was watched during the most active flying period. Seven males were caught in 24 hours. One was marked, and six were unmarked. All had flown at least three-fourths mile. It was noted that other males came to the cage from a direction against the wind but were not caught.

On August 13, 50 marked males were liberated at two points three-fourths mile to the southeast of the cage. The liberations were against the wind, as in the above experiment. The wind, however, soon shifted from northwest to westerly and southwesterly directions. No marked males were attracted, presumably owing to the shifting of the wind. Five unmarked males were caught in less than two hours. No marked males were caught in any of these experiments over four days after liberation. The nearest infestation was one-fourth to one-third mile distant.

On August 14 and 15, at 11.30 a. m., 75 marked males were liberated each day at three points three-fourths mile in an east-northeast to east-southeast direction from the cage. The wind was blowing from the west with a velocity of from 7 to 8 miles per hour. The temperature was 89° F. in the sun. A total of 33 males were caught, 6 of which were marked and had come against the wind. Two of them had been liberated at the ¾-mile liberation point August 11. The other four flew three-fourths mile against the wind and were caught after intervals varying from 55 minutes to 1 hour and 45 minutes from the time of liberation. Many were seen coming to the cage in general progress, but in a zigzag manner of flight, and were observed over a distance of 200 feet or more before reaching the female cage.

A series of liberations were made at Amherst, Mass., between July 25 and August 9, 1918, and repeated in July, 1919. In 1918 two cages, each containing 15 virgin females, were located on the campus of the Massachusetts Agricultural College, one 12 feet from the ground and the other 70 feet high on a building. It was hoped to compare the catch at the two heights and to learn whether males follow the scent directly or fly quite close to the ground regardless of the exact situation of the females.

A total of 914 males were liberated in 1918. No recoveries were made 70 feet above the ground, but three marked males were taken at the lower cage two days after being liberated, having come against the wind, apparently following the scent.

In the experiment in 1919 only the cage 12 feet from the ground was used. Marked males were liberated from the four cardinal directions, and out of 1,873 only 5 were recovered.

The direction of the wind during the interval between liberation and recovery indicated that the males sometimes came at right angles to the direction of the wind, or with it, as well as against it. The records also indicated from the small number of males recovered that they are ordinarily not attracted to females from distances of more than one-fourth or one-half mile. The distance they are actually attracted may be even less, as they apparently fly aimlessly for a considerable distance, finally locating the females by scent from shorter distances.

DISTANCES MALES FLY IN SEEKING FEMALES

When this method of attracting males is employed to locate infestations, it is important to know the maximum distances from which the males may come. To secure data on this phase of the problem some uninfested localities were selected on barren lands and rocky islands along the coast of Massachusetts and Maine, within a few miles of areas known to be infested. On selected sites cages were put out containing from 40 to 50 female gipsy-moth pupae from which 12 to 15 females issued. Fifteen treeless islands in Casco Bay and one off the coast of York, Me., were used for these experiments between August 3 and 25, 1915. These islands were situated at distances ranging from five-eighths mile to 7 miles from infestations with only the ocean between them. The catches of males ranged from 0 to 13 at each cage. Some scouting was done in the fall to determine the nearest known infestation on other islands. All the cages attracted males excepting two that were molested and one at Boon Island located 7 miles from the infested mainland. The largest catch consisted of 13 males, made on Upper Flag Island located five-eighths mile from an infestation on Haskel Island. Another catch of eight males was made on Little Bang Island. The nearest known infested island was Great Chebeague, five-eighths mile to the northwest. Another interesting catch consisted of four males on Sturtevant Island three-fourths mile northwest from the mainland, and the same distance from Basket Island to the south-southeast. The most significant catch was one of two males on Outer Green Island, located $2\frac{1}{12}$ miles from Jewell Island and $2\frac{3}{8}$ miles from Cliff Island, both to the northeast, and Peak's Island $2\frac{3}{4}$ miles to the northwest.

In 1917 some experiments on islands distant from infestations, and two additional experiments on the mainland, were repeated as checks. Two males were again caught on Outer Green Island and none at Boon Island as in 1915. Four males were caught on Billingsgate Island off Cape Cod, Mass., and scouting indicated that the nearest infestation was $2\frac{5}{16}$ miles north on the mainland. The males, in reaching this cage, were not obliged to fly more than a mile across water without resting, for a sandy point extends southward as part of the mainland.

These experiments indicated that small numbers of males can and do fly as far as $2\frac{5}{16}$ miles, but do not, under similar conditions, fly as far as $3\frac{1}{4}$ miles. Local conditions, such as those met with in these experiments, may increase the distance from a given point at which males may be recovered.

DISTANCES MALES FLY IN CONFINEMENT

In order to check the field experiments on distances males fly in seeking females, a few laboratory experiments were conducted in August, 1917. These were carried on in a room 12 by 14 feet, at a temperature of about 80° F., with curtains drawn to subdue the light. Preliminary tests indicated that males flew an average distance of 150 feet per minute where no air currents assisted or deterred their flight. Records were kept of the duration of flight and the number of times the males flew around or across the room, and distance computations were made from these. Males were used before and after mating. They are diurnal and were confined in large pill boxes at night.

Eleven males, 24 hours old, which had not mated, were tested first. One flew continuously for 1 hour and 2 minutes, and during another period 4 minutes. It covered a distance of 9,900 feet or 1.87 miles, and died the third day. The average flight for these 11 males was 0.62 mile. They all lived three days.

Ten males that were 48 or more hours old were allowed to mate. Two of these refused to fly, and two others flew only four and five minutes, respectively. The average flight of the eight males which flew was 0.66 mile. One of this lot flew continuously for 1 hour and 11 minutes, and the following day 13 minutes, covering 12,600 feet, or 2.38 miles. It flew 2 miles without resting. Specimens of this lot died the second, third, and fourth day after mating. Confinement in pill boxes undoubtedly hastened death. The indication that mated males fly slightly farther than unmated males may possibly be due to the small number of instances recorded.

It is interesting that the maximum distance covered by a male in confinement, namely, 2.38 miles, was very similar to the records secured on the treeless islands in Casco Bay near Portland, Me., and at Billingsgate Island, Mass.

TESTS OF TRAPPING MALES ON A LARGE SCALE

Since several striking catches of males had been made in border territory infested by the gipsy moth where it was not always possible to find an infestation within 1 or 1½ miles of the cage site, it was decided in 1917 to put out a large number of cages in border territory infested by the gipsy moth which would be subsequently scouted. Accordingly, 62 towns were selected adjacent to, but just inside, the 1917 quarantine line. This tier or strip of towns (double, or two towns wide, over much of the area traversed) extended from Long Island Sound, at the southeastern corner of Connecticut, to Southbridge and Monson, Mass., thence to Hinsdale, N. H., from which point it followed the Connecticut River Valley northward to Piermont, thence across to Chatham, N. H., which borders on the State of Maine. In the outer tier of towns the cages were three-fourths mile apart; in the inner tier they were placed 1 mile apart. An additional town, however, was selected where cages were placed 2 miles apart. A total of 2,743 cages were put out that year in the 62 towns, along 4,169 miles of road. There were from 19 to 177 cages per town, the number varying according to the size of the town[7] and the number of miles of road it contained. The cages were attached to the side of a tree away from the road (usually within 50 to 100 feet of it) and a "Do Not Molest" notice was attached to each cage. Fifteen carefully selected female pupae were placed in each double-walled cage, and a tin disk was tacked over the hole and sealed with sealing wax so that it would be known if any cages were molested. The average number of females emerging from the 15 pupae in the cages in three towns in Connecticut was 11.5, while the general average for the whole was somewhat less.

In 58 of the 2,743 cages put out that year no females issued, leaving a total of 2,685 cages charged with females. To these a total of 17,733 males were attracted, or an average of 6.6 males per cage. Many of the cages did not attract any males and many others only 1 or 2, while the maximum number caught at any one cage was 95. Not many more than this number would adhere ordinarily to the sticky surface of from 100 to 144 square inches, as the material is rendered ineffective by the scales from the wings and by the dead bodies of the males.

Table 1 shows that there were proportionately about as many males caught at cages placed 1 mile apart as at those three-fourths

---

[7] The area of towns in New England varies. The average size of the towns included in this experiment was approximately 38 square miles.

mile apart. This is because the inner tier of towns, where cages were placed 1 mile apart, was more heavily infested than the outer tier of towns, where cages were nearer together. It is thought that cages placed three-fourths mile apart gave results more indicative of the actual number of infestations in the towns.

TABLE 1.—*Average road miles and acres represented per male caught, 1917*

Distance between cages on road	Towns	Cages	Total road miles	Area [1]	Total males caught	Miles per male caught	Area per male caught [1]
	Number	*Number*	*Number*	*Acres*	*Number*		*Acres*
¾ mile	37	1,941	2,506.4	819,724	10,826	0.2315	77.55
1 mile	25	802	1,663.2	512,662	6,907	.2408	76.12
Total	62	2,743	4,169.6	1,332,386	17,733		

[1] The total area, and area per male caught, included in the table are based on the record for only 54 of the 62 towns (31 towns with cages three-fourths mile apart and 23 towns with cages 1 mile apart), as the exact acreage of 8 Connecticut towns is not known.

During the following fall and winter regular roadside scouting was done in many of the towns where cages attracted males, and data were compiled on the number and degree of the infestations found. In some cases it was impossible to determine whether the small number of males attracted to a given cage was due to the emergence of the females in the cage being much earlier or later than that of the males in the open infestation, or whether the attraction in the natural colony was great enough to prevent the males leaving their own locality. It is probable that the former is true, since in most cases where a fresh supply of females was introduced into cages near infestations the recovery was larger. After some scouting was done in the towns, records were compiled which indicated that catches at the cages in general compared with the number of egg clusters found at the nearest infestations. In New Hampshire and Vermont these infestations were found at an average of 1,813 feet (about one-third mile) from the cage, and those in Massachusetts and Connecticut 1,361 feet (about one-fourth mile) from the cage. These records further indicated that the males originated in the vicinity of the cage at which they were caught and did not fly long distances. Intensive scouting in the vicinity of some cages disclosed a few infestations not located by the scouts in their regular roadside work. It should be stated also that there were instances where infestations were found within a few hundred feet of cages that did not attract males. The records showed that the towns near the border were more heavily infested than previous scouting operations had indicated.

Some follow-up experiments were conducted in July, 1918, in 11 selected towns where fairly large catches of males were made in 1917. The catch was small in 1918. There were no males recovered at many cages, probably because of the extremely cold winter of 1917–18, which killed many gipsy-moth eggs and probably exterminated many isolated colonies. Some of the cages attracted from 1 to 12 males each in 1918, and during the spring of 1919 intensive scouting was done in a few selected localities where catches of 2 or more males had been obtained. Two reinfestations were discovered in Scotland, and one in Lisbon, Conn., towns which apparently had no infestations the preceding winter. Two new infestations and one reinfestation were found in Hardwick, Mass. All these infestations were less than one-eighth mile from points where cages that attracted males were located.

Similar results were obtained in 1919. Intensive scouting in December, 1919, revealed infestations 200 yards, and one-eighth, three-eighths, and one-half mile distant from cages that attracted males. In other instances old hatched egg clusters were found within a few hundred yards of cages that attracted males.

RECOVERY EXPERIMENTS WITH EXTRACTS IN MAINE

In 1928 an experiment was conducted in a very lightly or noninfested district at Dover-Foxcroft, near the center of the State of Maine, with the object of determining what percentage of males liberated in a given locality could be recovered by traps containing extracts of female abdominal tips (fig. 3) at various distances from the point of liberation. In addition, some information was desired on the influence of temperature, rain, humidity, topography, and forestation.

In natural infestations there is generally some counterattraction resulting from the scent of living females in the colony. Therefore, to approximate natural conditions, from three to five living unfertilized females were kept in cages from 3 to 10 feet from the points of liberation. Some males flew around and against these cages for a few minutes and soon left, while others would alight on a cage, remain for a while, and then fly away. Though the female seldom mates more than once, the males may mate from one to five times, if enough females are available. It also appears that more males normally emerge in a given infestation than females, owing to the fact that the shorter larval stage of the former insures a better chance for survival.

Two liberation experiments were made with 590 and 811 individuals, respectively. In each case an equal number of male pupae were put in rain-proof boxes attached to trees about 5 feet above the ground and so arranged that when the males issued they could fly away normally. The boxes were distributed at regular distances around a central extract trap. For the first experiment thirty 1-year-old tips in benzene were used for the attractant, and the liberations made at distances of ¼, ½, and 1 mile; and for the second experiment 15 tips of fresh material in xylene were used, and males were liberated at distances of 1/16, ⅛, and ¼ mile from the central trap. These distances are equivalent to the theoretical maximum distances of traps from road infestations along a straight road when traps are spaced twice these distances, for example, ⅛, ¼, ½, 1, and 2 miles apart. In the case of traps one-half mile apart, therefore, the greatest distance from an infestation along the road to the nearest trap will be one-quarter mile, and the average distance will be one-half this, or one-eighth mile. No marked males were caught at traps 1 mile and one-half mile from points of liberation, but 11, 17, and 69 males were caught at distances of ¼, ⅛, and 1/16 mile, respectively.

Upon the theoretical assumption that attraction varies inversely with the square of the distance—and if also it is limited to a distance of, say, one-fourth mile—then if a series of traps were distributed at half-mile intervals along a straight road, attraction would diminish to zero half way between the traps and in a circle one-fourth mile from each trap as a center.

The percentage of males that can be recovered by extracts is dependent on a number of variable factors, the more important of which are the distance they can fly, vitality and length of life, distance from the trap, number of males, wind direction and velocity, rain, humidity, temperature, counterattraction by females of a colony, tree growth, topography, and the attracting power of the extract. Considerable variation should be expected, therefore, in the results of the experiments.

The extracts were measured out at the laboratory at Melrose Highlands, Mass., and poured in 1-ounce vials stoppered with cork, and taken by automobile to Dover-Foxcroft. Unfortunately considerable of the attractive substance used in all the 1928 recovery experiments was lost by this method of handling, as is shown by the following comparative test: The contents of three vials brought back from Maine were put out at Saugus, Mass., on August 2, and the effectiveness of this material compared with that of six other traps containing xylene extract that had been taken direct from the Melrose Highlands laboratory to Saugus. The three traps containing unused material brought from the Maine recovery experiments caught an average of 22.3 moths, while the six traps of fresh laboratory material caught an average of 64.7 moths.

The low summer temperature in the locality where the Maine experiments were conducted was one of the main causes for the low recovery at the traps. Observations, both at the laboratory and in the field, showed that the males would never fly voluntarily when the temperature was below 70° F., and that their activity increased as the temperature rose from 70° to 90°. One observation was made at 95°, when activity was at least as great as at 90°. The number of males caught varied with the number of available hours [8] and with the number of degrees of cumulative excess temperature [9] above 69°. (Table 2.) The distance males can fly to extract traps depends considerably on the temperature. At Dover-Foxcroft, Me., there were only 60 per cent as many hours when the temperature was above 69° as at Melrose Highlands, Mass., and only 33 per cent as much cumulative temperature above 69°. Rain or wet foliage stopped the flight of males, but they appeared to be attracted somewhat better when the heat was accompanied by humidity, provided the foliage was dry.

---

[8] An available hour for flight of males is an hour during which the temperature is above 69° F.

[9] A degree of cumulative excess temperature is counted by each degree of temperature above 69° F. during an hour. A temperature of 73° F. for one hour would make 73 – 69 or 4 degrees of cumulative excess temperature. The same for three hours would give 12 degrees of cumulative excess temperature, etc.

TABLE 2.—*Effect of certain weather conditions on recovery of males of the gipsy moth, 1928*

FIRST LIBERATION—FIRST SERIES OF EXPERIMENTS

Date of liberation	Dover-Foxcroft, Me. Hours above 69° F.	Dover-Foxcroft, Me. Cumulative temperature	Dover-Foxcroft, Me. Males caught	Dover-Foxcroft, Me. Character of day	Melrose Highlands, Mass. Hours above 69° F.	Melrose Highlands, Mass. Cumulative temperature
	Number	*°F.*	*Number*		*Number*	*°F.*
July 20	0	0	0	Clear	0	0
July 21	10	39	2	Clear, calm	9	29
July 22	6	10	0	Cloudy	0	0
July 23	0	0	0	Rain	2	2
July 24	8	19	2	Clear	7	18
July 25	12	60	2	Clear, warm	10	111
July 26	6	8	1	Clear, calm	4	4
July 27	0	0	0	Cloudy, rain	8	16
July 28	0	0	0	Heavy rain	7	18
July 29	0	0	0	Clear	9	9
July 30	0	0	0	do	7	13
July 31	7	14	0	do	8	41
Total	49	150	7		71	261

SECOND LIBERATION—SECOND SERIES OF EXPERIMENTS

Date of liberation	Dover-Foxcroft, Me. Hours above 69° F.	Dover-Foxcroft, Me. Cumulative temperature	Dover-Foxcroft, Me. Males caught	Dover-Foxcroft, Me. Character of day	Melrose Highlands, Mass. Hours above 69° F.	Melrose Highlands, Mass. Cumulative temperature
August 1	10	52	21	Partly cloudy, warm	10	40
August 2	8	36	25	do	12	148
August 3	5	4	12	Partly cloudy to showers	14	184
August 4	9	52	24	Clear to partly cloudy	14	196
August 5	0	0	0	Cloudy	12	52
August 6	0	0	0	do	0	9
August 7	3	8	6	do	0	0
August 8	0	0	0	Rain	0	0
August 9	0	0	0	do	6	80
August 10	1	1	1	Cloudy	1	18
August 11	1	1	1	Clear	4	1
August 12	0	0	0	do	0	1
Total	37	154	90		73	666
Total for both liberations	86	304	97		144	927

When liberated in an open field the males showed a very noticeable tendency to seek the edges of woods and roadsides, many entering the woods after flying back and forth along the edge. It was also noticed that males in the woods very seldom come out into open fields unless attracted outside by the scent of females. Whether this tendency to fly in the vicinity of tree growth is instinctive, or arises from the use of sight, or from the detection of forest odors is a question.

Where there are hills between the point of liberation and the trap, not so many males are caught as where the males have unobstructed fields to fly across.

CHEMICAL STUDIES OF THE ATTRACTING SUBSTANCE

Early experiments indicated that the males were attracted by an odor given off by the females. It was hoped that it would be possible to determine whether a definite substance of attractive power was present in the females, and if so to isolate it, determine its nature, and, if possible, prepare it in quantity synthetically for use in trapping the moths. Accordingly, A. F. Burgess and the senior author approached W. R. Bloor, of the Harvard Medical School, and engaged him temporarily to assist in making these studies. Doctor Bloor's report[10] is in part as follows:

In a large cage out of doors, gipsy moth males were strongly attracted, not only to living females but also to the whole or parts of the abdomen of females. When males had begun to fly in considerable numbers in Sharon, Mass., most of the succeeding experiments were conducted at that place. Later, when moths were present in considerable numbers at the Medical School in Boston, experiments were conducted there.

The work was made quantitative by use of the "Shaw" trap[11] covered with a sticky mixture. The trapping tests were carried out along two lines; (1) with various extracts of the female moth, and (2) with chemical substances, and later with natural products, e. g., resins. As regards the second type of substances (chemicals, etc.), many were tried, and the following attracted moths in the order named: Allylamine, ethylamine, and methylamine. Slightly active were quinoline, which attracted very many, but caught few (three in one-half hour), and benzonitril which caught several flies but only one moth. A peculiarity of all of these substances was that they attracted when first put out but were not sufficiently attractive to catch a large number of moths, and lost their power in the course of an hour or so, pointing to the probability that there were present traces of an actively attracting substance which was soon lost by volatilization.

Results from extracts made from the bodies of female moths may be summed up as follows: (1) Water extracts contained only traces of the active substance, and this could be extracted from the water by shaking with ether. (2) Ninety-five per cent alcohol extracted all the active substance. (3) The active substance was readily soluble in ether and less readily soluble in petroleum ether (naphtha). (4) These solubilities indicated that the substance was probably a lipoid (fat or related substance). A sample was saponified with sodium ethylate and a separation made of saponifiable (fatty acids) from unsaponifiable substances. In this "unsaponifiable" group would be included various substances belonging to the aromatic group, as cholesterol, resins, terpene derivatives, etc. Both saponifiable and unsaponifiable parts were tested for their power of attraction, and the active principle was found to be present almost entirely in the unsaponifiable portion. Attempts to divide this portion by fractional distillation at low pressure with temperatures up to 100° C. and with the receiving bulb in a freezing mixture were not successful. The crude petroleum ether extracts were fractioned as regards their solubility, and the active substance was found to be in the most soluble fraction. (5) Extracts made from various portions of the body of the female showed that the attractive substance was present in quantity only toward the last segment of the abdomen; and, therefore, probably had its origin in the accessory sexual glands of the oviduct. (6) The active substance seemed to be quite stable. The first alcoholic extract of abdomen, free from eggs and hair, remained active 22 days when exposed in the field. When freed from its fatty impurities it was active for a shorter time, owing possibly to its volatilization not being impeded by these fatty substances. (7) The males seemed much more sensitive to attraction in the fore part of the season than in the latter part. They would at first be attracted to extracts containing probably traces of the active substance, which later in the season had no effect on them.

The results thus far obtained appear to justify the following conclusions: (1) That the males are attracted to the females by the odor of a substance produced by the females, probably in glands toward the last segment of the abdomen, and (2) the substance is relatively stable, is soluble in alcohol and in the ordinary "fat solvents," and slightly in water. After saponification the active material appears in the unsaponifiable fraction.

---

[10] Unpublished.

[11] The Shaw trap has four vertical wings like a 4-bladed paddle wheel set on end, with a small cage for the females set into an opening made where the blades intersect at the bottom. These blades are covered with a sticky mixture which catches and holds the insects. The insects coming from any certain source are naturally caught in the angle opening in that direction.

The next season C. H. Fiske, biological chemist at the Harvard Medical School, was temporarily engaged. A part of his report[12] is as follows:

Proceeding on the assumption that the active material is volatile, an examination of distillates from alcoholic extracts of female abdominal tips was made along various lines.

Analysis of one such extract (1,500 abdominal tips) showed it to contain 35 milligrams of nitrogen. A portion of the same extract, distilled from alkaline solution, gave an amount of volatile base (in the distillate) equivalent to 33 milligrams of nitrogen. This distillate, acidified with hydrochloric acid and evaporated to dryness, left a small residue consisting, as far as could be determined, entirely of ammonium chloride. Practically all the nitrogen in the original extract was, therefore, recovered as ammonia, leaving no possibility of the presence of other volatile nitrogenous bases in more than the minutest traces. The chances of the active substance being of this nature is consequently very slight.

In the course of the above procedure, it was found that the distillate gave a bulky yellow precipitate (mercury compound) with Nessler's reagent (an alkaline solution of mercuric potassium iodide). The same yellow substance was obtained in purer condition (since uncontaminated with ammonia) after distillation of the alcoholic extract with acid instead of alkali. It contains 66 per cent of mercury and gives qualitative tests for iodine, but is free from nitrogen, sulphur, and phosphorus. After removing the mercury from the precipitate, and distilling again, this time from alkaline solution, there is still about the same quantity of substance concerned to be found in the distillate, showing (1) that it can be recovered from the mercury compound, and (2) that it is not an acid.

Similar precipitates were obtained from other mercury reagents (reddish precipitate with mercuric chloride; white precipitate with an alkaline solution of mercuric cyanide). These tests suggest the presence of an aldehyde or a ketone, and it was accordingly found that the distillates gave a strongly positive test for aldehyde with Schiff's fuchsin reagent (red color). Analysis of the above distillate by Ripper's method showed the presence of 0.03 per cent aldehyde (calculated as acetaldehyde). By the same method, the distillate from another extract containing a larger number of tips showed 0.06 per cent aldehyde.

Attention was then directed toward the characterization of the aldehyde. The most suitable reagents for this purpose were not available at the time, so as much information as possible was sought with reagents that were in stock or that could be prepared readily. After various unsuccessful preliminary tests with other reagents, thiosemicarbazide was finally selected as suitable for more detailed investigation, since both it and its aldehyde derivatives are readily analyzed, particularly in the form of their silver compounds. Nevertheless, in spite of numerous attempts to isolate a compound of this reagent with the aldehyde present in the distillates, nothing has so far been recovered but a substance which, as shown by its physical properties and by the silver content of its silver compounds, is merely unchanged thiosemicarbazide. The desired condensation product, therefore, either is not readily synthesized under the conditions adopted, or is too unstable to resist the manipulations that would be necessary to isolate it.

The most promising direction for further work would be an attempt to apply, in a similar way, other reagents not now obtainable on the market, particularly p-nitrophenyl-hydrazine.

When the services of Doctors Bloor and Fiske were no longer available, B. L. Souther, organic chemist working at Harvard University, was temporarily engaged to assist in making the chemical investigations. E. P. Kohler, head of the department of organic chemistry at Harvard, directed the work of Doctor Souther and offered many suggestions for trial and improvement. A summary of Doctor Souther's report[12] is as follows:

---

[12] Unpublished.

Experiments were first made with old material held over winter in cold storage (0° to 3° C.) and later checked with fresh material with the object of determining (1) the general class, or classes, of organic compounds to which the attractive substance belongs, (2) the procedure that would give the most complete extraction of the material from the abdominal tips, and (3) a possible method of isolating the substance. The samples were put out in tin evaporating boxes and attached to trees.

Thirty-two samples of filtered extract, each from a minimum of 250 abdominal tips in 21 c c of petroleum ether, were treated with various reagents and then subjected to field tests, with the results recorded in Table 3. Four untreated samples of the same concentration were used as checks. All of the samples were prepared from female tips. The more important experimental procedure is described below.

TABLE 3.—*Tests with samples of the solution of abdominal tips of female gipsy moths, showing numbers of male moths attracted to traps after various treatments of the solution, Saugus, Mass., 1921*

Sample No.	Treatment of sample [1]	Female tips used	Total males caught	Sample No.	Treatment of sample [1]	Female tips used	Total males caught
		Number	*Number*			*Number*	*Number*
1	Check solution [2]	250	95	17	Distillate evaporated on filter paper	250	3
2	Treated with sodium carbonate to remove acids	250	374	18	Test for base as in sample 3	500	13
3	Treated with sulphuric acid to remove bases	250	117	19	Hydrolysis of extract with potash	250	15
4	Treated for unsaturated compounds	250	13	20	Test for unsaturated compounds	250	183
5	Test for aldehyde	1,000	119	21	Test for aldehyde	250	182
6	Oxidation of dried tips	1,000	7	22	Boiled with potash	250	86
7	Hydrolysis of dried tips	1,000	119	23	Most volatile of two portions	250	8
8	Most volatile third of three lots	250	217	24	Second portion of distillate	250	75
9	Second portion of three lots	250	73	25	Solvent added and distilled off	250	41
10	Third portion of three lots	250	16	26	Distilled at 150° C	250	207
11	Residue distilled at 100° C	250	1	27	Solvent added and distilled off	250	23
12	Residue distilled at 125° C	250	0	28	Residue	250	8
13	Residue distilled at 150° C	250	314	29	Check solution [2]	250	214
14	Final residue	250	30	30, 31, 32, 33	See text	- 250	1
15	Check solution [2]	250	118	34	Hydrolysis of extract with sodium carbonate	250	42
16	Nonvolatile portion of one lot	250	15	35	Check solution [2]	250	8
				36	See text	250	0

[1] Refer to text for more detailed information on the composition and treatment of each sample.
[2] Check sample 1 covers samples 1 to 14, check sample 15 covers samples 15 to 28, check sample 29 covers samples 29 to 34, and check sample 35 covers sample 36.
[3] 250 used in each sample.

TEST FOR ACID

An old lot was extracted twice with 5 per cent sodium carbonate solution, washed with water, and dried. (Sample 2.) This sample attracted more moths than the check solution, proving a previous conclusion that the substance is not an acid. A new lot was also hydrolyzed with sodium carbonate and caught 42 moths. (Sample 34.)

The greater attractiveness or activity of sample 2 as compared with the check solution indicates that the active material is continuously being generated by hydrolysis from a more complex compound.

139621—32——3

A sample of fresh material was hydrolyzed with the more active potassium hydroxide, instead of sodium carbonate, and the extract was not very active, indicating that it was not stable in strong, cold alkalies. (Sample 19.)

HYDROLYSIS WITH ACID

One thousand tips which had been covered all winter with petroleum ether were decanted and washed with petroleum ether and dried in the open air. As soon as the tips became dry they were moistened with 1 per cent hydrochloric acid and shaken overnight. The acid was neutralized with sodium carbonate, and the resulting solution extracted with ether. (Sample 7.) It caught many moths, thus confirming the hypothesis that hydrolysis plays an important rôle in causing extracts to retain their activity, the moisture of the air acting as the hydrolyzing agent in the field experiments. Oxidation is not concerned in the process, as shown by the results with 1,000 tips which had been decanted, washed with ether, and dried, as in sample 7. These were moistened with water and shaken with oxygen under 13 pounds pressure, the material extracted with ether, and the solution dried, but when tested it caught only seven moths. (Sample 6.)

TEST FOR BASE

A sample was extracted twice with 5 per cent sulphuric acid, washed with water, and dried. It caught many moths; this shows that the attractive material is not a base. (Sample 3.) Six per cent hydrochloric acid was next used instead of sulphuric acid, but this sample was only slightly active. (Sample 18.)

TEST FOR UNSATURATED COMPOUNDS

Potassium permanganate in neutral solution readily oxidizes many unsaturated compounds. Therefore, two samples were shaken overnight, each with 25 c c of a 3 per cent solution of potassium permanganate. (Samples 4 and 19.) The results showed conclusively that the attractive principle is a saturated compound and is not unsaturated.

TEST FOR ALDEHYDES

A portion of extract was boiled for two hours with hydroxylamine and potassium hydroxide. The alcoholic solution was evaporated and the residue poured into water and extracted with ether after acidification with weak hydrochloric acid. This procedure would remove all aldehydes and all but complex ketones. The extract attracted many moths; this shows that the active principle is not an aldehyde. (Sample 5.) The test was repeated, and at the same time another extract sample was boiled with potassium hydroxide in order to determine whether the alkali or the hydroxylamine caused the change, should any be observed in the solution. Both samples were active. (Samples 21 and 22.)

VOLATILITY TESTS

It was also desirable to determine, if possible, the volatility of the attractive constituent. Accordingly, a quantity of the solution equivalent to three lots (63 c c) was fractionally distilled from a steam bath. The first 21 c c that distilled was very attractive and caught 217 moths. (Sample 8.) The second fraction that came over was less attractive and caught 73 moths. (Sample 9.) The third distillate consisted of the last portion that would not distill at steam-bath temperature, and was even less attractive, catching only 16 moths. (Sample 10.) The residue was heated for an hour at 100° C. in a current of air under diminished pressure, but the distillate was not active. (Sample 11.) The heating was continued at 125° for an hour; yet no active material distilled. (Sample 12.) After an hour's heating at 150° a small amount of distillate collected, and this sample was very active. (Sample 13.) The discolored residue that remained in the flask was washed out with solvent and tested, but it caught very few moths. (Sample 14.)

A lot was evaporated in a vacuum at room temperature, the distillate being collected in a flask immersed in a freezing mixture. The portion that would not distill caught very few moths. (Sample 16.) To present as large a surface as possible for evaporation, pieces of filter paper were put in the flask containing the condensate. The liquid was then evaporated under diminished pressure. The paper and the small amount of liquid which remained in the flask caught very few moths. (Sample 17.) That few moths were caught from these two tests (samples 16 and 17) is very strange, but local field conditions may have been responsible for the result.

Another series of volatility tests were carried out using a double portion of the extract. The most volatile portion was only slightly active (sample 23), but the second portion of the distillate showed considerable activity. (Sample 24.) In order to remove, if possible, all of the active compound, petroleum ether was added to the residue and distilled off immediately. This solution caught many moths, but quickly lost its power of attraction. (Sample 25.)

These experiments indicate that the active compound is readily volatile with solvents. One seems justified in supposing, therefore, that in field tests where a preliminary evaporation of the solvent takes place, a great loss of the active material is incurred.

THERMAL TEST

In order to further determine whether the attractive substance could be obtained by heating the residue similar to that left behind in sample 25, it was heated to 150° C., no air current being used. The small amount of distillate contained much of the active substance. (Sample 26.) As no air current was used during the heating, it was hoped that if the active material were generated in the flask, it would remain there and could be obtained by adding petroleum ether and distilling it off again. The distillate made a good catch on the first day only. (Sample 27.) The final residue was inactive. (Sample 28.)

Identical original samples (30, 31, 32, and 33) were each shaken with 5 c c of concentrated hydrochloric acid overnight. The aqueous portion was distilled, made faintly alkaline with potassium hydroxide, and extracted with ether. The four samples caught only one moth, which seems conclusive evidence that the active material is destroyed by the action of concentrated hydrochloric acid.

By the time these experiments had been completed, tips were available in sufficient quantity to attempt to isolate the active compound. The most promising means to this end seemed to be fractional distillation, since the chemical nature could not be determined.

Accordingly, a filtered solution of 1,650 c c from 21,625 moths was evaporated under diminished pressure with the receiver in a freezing mixture. The last portion came over at 40° C. under a pressure of about 6 inches of mercury. The residue was distilled, and the portion that would not volatilize at 175° and 20 mm pressure was dissolved in ether and hydrolyzed with sodium carbonate solution. The ether layer was washed and evaporated and the residue steam distilled. The aqueous distillate was extracted with ether. The ether was then evaporated and residue tested. (Sample 36.) It caught no moths. It seems that the substance is either volatilized or destroyed at 175°.

One may briefly conclude, therefore, from the foregoing experiments that the attractive principle is not an acid, base, or aldehyde. It is probably an indifferent substance, as a fat, protein, or ester, and is saturated.

Cold concentrated hydrochloric acid and boiling alcoholic potash destroy the substance.

It is readily volatile with solvents, and much is lost when the solvent is removed by evaporation before the sample is placed in the field.

The long activity of extracts is probably due to the continuous generation of the attractive compound by the hydrolysis of a more complex substance. Oxidation is not concerned in the process.

Abdominal tips that have been thoroughly extracted with petroleum ether yield an additional amount of active material on hydrolysis with 1 per cent hydrochloric acid.

The samples were put out between July 21 and August 1 and remained exposed in the field for a period of 25 to 14 days. In Table 3 they are listed in the order in which they were put out. A catch of five males, or less, may be considered as negative. The catches were recorded daily, and from these the totals given in the table were obtained.

ANATOMICAL STUDIES

Studies made in 1927, 1928, and 1929 were planned (1) to get more definite information on how much of the female abdomen should be clipped into the solvents for best results, (2) to determine exactly what part of the anatomy produced the attractive substance, and (3) to obtain information which might aid in determining the exact nature and chemical formula of the attracting substance so that it might be prepared in quantity synthetically.

Various parts of the anatomy, more particularly the reproductive system, were dissected, then placed in a solvent (xylene), and put out in traps in the field. Most of the dissecting was done under distilled water, in which previous tests had shown a small portion of the active substance to be soluble. Therefore, while making the dissections, a small quantity of the active substance became absorbed by the various parts of the anatomy as they were being removed. Hence, a small number of male moths were sometimes caught by these parts (particularly in 1927) in traps which otherwise would not have caught any males. These traps were put out in heavy infestations in 1927 and in moderate infestations in 1928 and 1929. Under these conditions a catch of 12 males or less may be considered as a negative result. After 1927 it was found that considerable dissecting could be done without a liquid; but when it became necessary to dissect certain parts under water, this was done as quickly as possible. In 1927 only a relatively small number of parts were dissected, and each sample was held in one-fourth ounce of xylene for approximately 5 days and then put in the field. In 1928 and 1929 each sample of parts was put in three-fourths ounce of xylene for approximately 10 days before being put in the field. During these two years several hundred females were dissected, so that duplicate tests of each sample of parts were made, and it is mostly from these tests that the conclusions have been drawn, instead of from the preliminary results of 1927.

TABLE 4.—*Numbers of male gipsy moths caught at traps containing dissected parts of females, 1927* [1]

Sample No.	Females used	Part of female used	Males caught	Males caught per female represented
	Number		*Number*	*Number*
1	1	Tissue at oviduct opening	82	82
2	1	Tissue immediately surrounding opening of copulatory pouch	98	98
3	3	Copulatory pouch	2	0.7
4	3	Oviduct	36	12
5	4	Accessory glands	9	2.2
6	12	Ovarian tubes	51	4.2
7	2	Rectal pouch	2	1
8	12	do	41	3.4
9	10	Liquid from rectal pouch	3	0.3
10	1	First segment (from posterior end, no part of reproductive system included).	8	8
11	15	"Tips" ground in 5 cc water, plus 1 ounce xylene	507	33.8
12	15	Large female larvae ground in 1 ounce xylene	0	0
13	15	Female pupae ground in 1 ounce xylene	0	0
14	15	Head and thorax of female moths	0	0
15	25	Abdominal "tips" alone (no solution)	5	.2
16	30	Check (living females)	848	28.2

[1] The traps were placed in the field on Aug. 1, where they remained 3[illegible] days.

It has been assumed by many zoologists that the substance which attracts males to females is produced in the accessory glands of the reproductive system. In the reproductive system of the female gipsy moth there are two accessory glands: One a small bulblike structure whose function is not known unless it is that of a seminal vesicle; and the other a large paired gland containing the substance which, upon exposure to air, cements the eggs together and to the surface on which they are deposited and which, unlike the attractive substance, does not mix or dissolve in fat solvents. The contents of both of these glands gave negative tests for the attracting substance. It is interesting that tests with the reproductive system (the accessory glands, copulatory pouch, oviduct, ovarian tubes, and eggs) yielded generally negative results, as likewise did traps containing the hairs, rectal pouch, segments of the abdomen taken after removal of the reproductive system, the thorax and head of the adult, and female larvae and pupae. The data presented in Tables 4, 5, and 6 indicate that the attractive substance occurs in the tissue immediately surrounding the opening of the oviduct and copulatory pouch, more particularly in the vicinity of the copulatory pouch opening. This tissue consisted of the genitalia, together with a very thin, narrow band of body-wall tissue of about one-half millimeter in width. In dissecting these chitinous parts it was found that often a very small quantity of tissue that was attached to the inner chitinous surface was also removed. In most instances that portion of the genitalia pertaining to the copulatory pouch opening and that portion surrounding the oviduct opening were put out in separate traps.

TABLE 5.—*Numbers of male gipsy moths caught at traps containing dissected parts of females, 1928*

Contents of traps	First series of tests			Second series of tests		
	Females used	Days exposed in field	Males caught	Females used	Days exposed in field	Males caught
	Number	*Number*	*Number*	*Number*	*Number*	*Number*
Tissue immediately surrounding copulatory and oviduct opening	19	5	[1] 13	15	10	34
Tissue immediately surrounding copulatory opening				15	10	14
Tissue immediately surrounding oviduct opening				15	10	20
Copulatory pouch	18	15	0	15	10	0
Oviduct	17	15	6	15	10	0
Accessory gland (small bulblike)	17	15	0	15	10	0
Accessory glands (large paired)	19	15	0	14	10	0
Ovarian tubes with approximately 4,500 eggs	21	15	4	16	10	1
Eggs (approximately 4,500) alone	14	15	0	17	10	0
Rectal pouch	18	15	0	14	10	1
Liquid from rectal pouch	15	30	1	16	10	0
Dissecting water shaken with xylene	18	14	29	13	10	25
Connective tissues—thorax, etc	14	15	8	14	10	4
Hairs from abdomen	30	26	3	15	10	1
First segment from posterior end (without reproductive system)	12	14	3	14	10	5
Second segment from posterior end (without reproductive system	14	14	1	14	10	0
Third segment from posterior end (without reproductive system)	12	15	[2] 40	15	10	0
Larvae (ground)	16	15	0	16	10	0
Female pupae	13	15	3	17	10	0
Dead females about 14 days old, crushed	18	15	0	18	10	0
Blank test (tree-banding material only)	0	15	0	0	10	0
Check (living virgin females)	30	15	170	25	10	144

[1] Trap molested after fifth day.
[2] Female found ovipositing on tree under trap, which probably accounts for the results.

TABLE 6.—*Numbers of male gipsy moths caught at traps containing dissected parts of females, 1929*

Contents of traps	First series of tests [1]		Second series of tests [2]	
	Females used	Males caught	Females used	Males caught
	Number	*Number*	*Number*	*Number*
Oviduct	18	5	15	1
Copulatory pouch	16	3	12	1
Small bulblike accessory gland	19	18	15	0
Large paired accessory glands	18	1	15	1
Ovarian tubes with eggs	18	7	15	1
Tissue around oviduct opening	19	101	15	53
Tissue around copulatory pouch opening	18	101	15	137
Tissue near opening of oviduct and copulatory pouch			15	12
First segment from posterior end (without reproductive system)	16	25	15	[3] 30
Do			15	2
Second to fifth segments from posterior end (without reproductive system)	18	7	15	[3] 62
Do			15	4
Dissecting water (130 c c in xylene)	20	40	15	48
Living females (check)			16	716

[1] Traps placed in field July 26, where they remained 29 days.
[2] Traps placed in field July 30, where they remained 26 days.
[3] Results questionable, for most of the other tests of these parts gave negative results, and because these traps attracted males on the first day only. Furthermore, in 1930 a trap containing the genitalia of 15 females in xylene caught 279 males, while the rest of the abdomen of these females put out in the same way caught only 20 males.

Since the chemical tests indicated that the attractive material may arise from some fatty substance, it seemed advisable to trace roughly the metabolism of fat bodies in the larva, pupa, and adult. The healthy female larva at its fullest development has an internal cylinder of fat which surrounds the alimentary canal and lines the body cavity. In the pupal stage the fat bodies diminish as the reproductive system and eggs develop; the development of the latter taking place at the expense of the former. There is very little fat left in the body of the adult female, but there is an appreciable amount in the male. As the male is very active, it is possible that this fat may be necessary to furnish the energy expended in flight.

In conclusion, the field experiments with dissected parts strongly indicate that the active substance is not produced internally by glands in the reproductive system. Since the tissue of the reproductive system apparently is not the kind of tissue which contains fatty substances, and since previous chemical tests indicated that the substance is of a fatty nature and is soluble in fat solvents, it seems quite evident that the attractive substance is produced elsewhere. These experiments, covering a period of three years, indicate that it comes from tissue immediately surrounding the copulatory pouch. Here, it may arise from the decomposition and hydrolysis of a fatty substance. It is not unlikely that it is produced and excreted in this vicinity by ductless or hypodermal glands lying near the surface, especially since setae and pores were found here that are peculiar to this region.[14]

TESTS OF EXTRACTS

EXPERIMENTS FROM 1920 TO 1927

The principal results, up to 1927, with extracts made from female abdominal tips are summarized in Table 7. The table contains results taken from 322 traps at Saugus, Mass., covering a period of several years. The total number of males caught by each extract was obtained by adding the daily catch for the entire season. All traps, after being put out, were visited until there were no longer living males in the field. The period over which the traps were exposed in the field is important because of its bearing on the number of moths caught. Those that were exposed longest were put out

---

[14] In reviewing his own work and the work of others, McIndoo[15] wrote the following: "* * * Gazagnaire (1886) remarks that glandular cells of hypodermal origin are widely distributed in insects. They secrete the various fluids exuding through the chitin, and since their histology is so similar it might be admitted that they have the same general structure. For description, scent-producing organs may be divided into five types based on their devices for disseminating the odor and for storing the secretion as follows: (1) No special device for disseminating the odor or storing the secretion; (2) gland cells associated with hairs and scales as a means of scattering the odor more effectively; (3) "evaginable" sacs lined with hairs connected with gland cells as a device for storing the secretion and distributing the odor; (4) articular membranes serving as pouches for storing and preventing a too rapid evaporation of the secretion; (5) specialized tubes and sacs acting as reservoirs for storing and discharging the secretion." This subject is further treated by McIndoo in "Smell and Taste and Their Applications"[16], and "Communication Among Insects."[17]

[15] McIndoo, N. E. RECOGNITION AMONG INSECTS. Smithsn. Misc. Collect. 68, no. 2, p. 65. 1917.

[16] ——— SMELL AND TASTE AND THEIR APPLICATIONS. Sci. Mo. 25: 481–503, illus. 1927.

[17] ——— COMMUNICATION AMONG INSECTS. Smithsn. Inst. Ann. Rpt. 1928: 541–562, illus. 1929.

earlier than the others, and these generally caught more males than those put out later because (1) they were exposed for a longer period of time, and (2) because the flight of males reaches its peak early in the season. This condition is illustrated by experiment 1, Figure 4. This experiment, using living females as the source of attraction, was started at Saugus, Mass., July 10, 1921, and lasted 41 days. The peak, for males caught, was reached in the fourteenth day. The seasons vary somewhat from year to year, but usually the first males start issuing about July 10, and the females a few days later. An occasional male may be found as late as September 1, in the vicinity of Melrose Highlands, Mass., and later in northern New England, making a season six to eight weeks in length.

TABLE 7.—*Summary of data on experiments for trapping male gipsy moths using extracts from abdominal tips of females, 1920–1927*

Solvent	Female tips used	Age of extract	Year of test	Traps used	Days in field	Males caught
	Average	*Years*		*Number*	*Average*	*Average*
Anhydrous ether	125.9	[1] 0	1920	10	36	145.4
	348	1	1920	10	36	76
	100	2	1920	1	36	1
	25	0	1921	1	11	36
	87.5	1	1922	2	37	147
Petroleum ether	405	0	1920	9	26	84
	202	1	1920	1	36	0
	107	3	1920	1	36	2
	30	0	1921	11	28.5	141
	135	1	1922	2	37	240
	25	0	1922	1	33	127
	125	1	1923	1	37	14
	1,000	3	1923	1	37	1
Wood alcohol	25	0	1921	1	31	113
	55	1	1922	1	36	162
Ethyl alcohol 95 per cent	225	1	1921	1	40	384
	200	2	1921	1	40	240
	25	0	1921	3	39	190
Alcohol on bark	25	0	1921	2	25	213
Alcohol on cotton	25	0	1921	2	25	121.5
	122.5	1	1922	2	37	340
Alcohol on bark	25	0	1922	1	29	121
Alcohol on cotton	500	5	1923	1	30	0
	64	5	1927	1	40	671
	250	7	1927	1	40	13
	500	7	1927	1	40	397
Chloroform	25	0	1921	1	39	4
Carbon tetrachloride	25	0	1921	1	32	160
	25	2	1927	1	40	65
Benzene	25	0	1921	7	28	84
	101	1	1922	3	37	325
	25	0	1922	2	22	85
	18	0	1923	18	28	25
	13	1	1924	11	37	20
	15	0	1924	3	30	38
	30	0	1925	7	48	56
	17	0	1926	5	37	247
	30	2	1927	1	43	735
	30	1	1927	2	43	442
	18	0	1927	2	43	405
Benzene plus 10 drops tincture of civet	15	0	1927	1	43	611
Benzene and gasoline [2]	50	1	1923	1	38	87
	12	0	1923	3	38	29
Benzene and vaseline	15	0	1925	7	44	1
	30	1	1925	4	44	36
Gasoline	26	1	1923	2	38	40
	17	0	1923	10	38	35
	8	1	1924	11	38	16
	15	0	1924	2	29	13

[1] Zero indicates fresh material. [2] Cheap grade of gasoline used in all experiments.

TABLE 7.—*Summary of data on experiments for trapping male gipsy moths using extracts from abdominal tips of females, 1920–1927*—Continued

Solvent	Female tips used	Age of extract	Year of test	Traps used	Days in field	Males caught
	Average	*Years*		*Number*	*Average*	*Average*
Xylene	20	1	1924	2	37	34.5
	15	0	1924	1	30	53
	30	1	1925	1	44	123
	15	0	1925	2	31	13.5
	17	0	1926	7	38	304
	30	1	1927	2	44	323
	16	0	1927	9	30	242
Geraniol	15	0	1927	2	36	1
Tincture of musk	0	0	1927	2	18	0
Tincture of civet	0	0	1927	2	38	54.5
Commercial vegetable oils	0	0	1927	4	36	0

In studying and comparing results, several natural conditions must be taken into account, the most important of which are (1) the degree of infestation where experiments were conducted as indicated by the checks (Table 8) and (2) the portion of the season over which the experiment is made. Extreme variations may also occur from day to day, owing to weather conditions. Cool rains, wet foliage, and temperature below 70° F. entirely stop the activity of the males.

TABLE 8.—*Average catch of males at check traps containing living virgin females, Saugus, Mass., 1920–1930*

Year	Cages	Average catch of males	Year	Cages	Average catch of males	Year	Cages	Average catch of males
	Number	*Number*		*Number*	*Number*		*Number*	*Number*
1920	2	500	1924	2	301	1928	4	335
1921	2	550	1925	1	958	1929	3	318
1922	1	1,513	1926	2	1,135	1930	1	395
1923	1	976	1927	2	1,085			

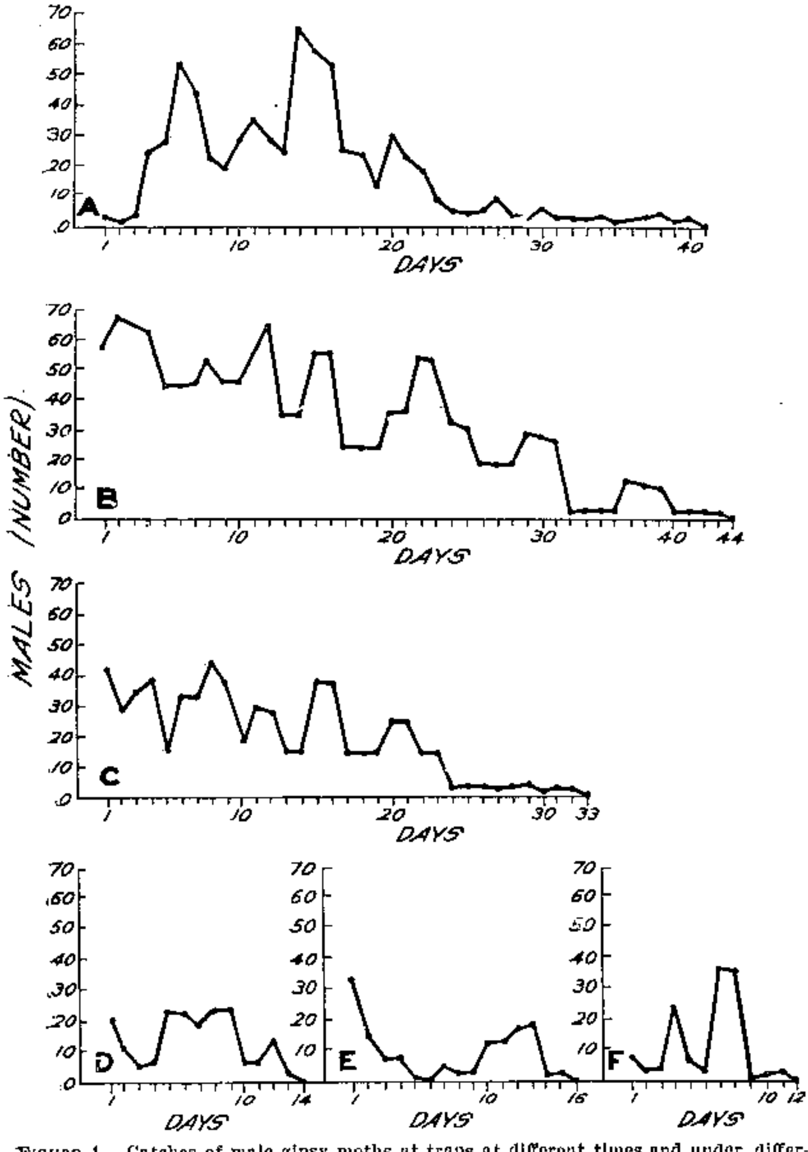


FIGURE 4.—Catches of male gipsy moths at traps at different times and under different conditions: A, Experiment 1, check, using 40 living females, July 10 to August 19, 1921, showing the beginning, height, and end of catch; B, experiment 2, check, using 40 living females, July 25 to September 6, 1927, indicating the relative abundance of males from July 25 to the end of the season; C, experiment 3, average catch of two traps containing 30 tips extracted with benzene under the same conditions as in experiment 2; D, experiment 4, petroleum ether extract of 15 tips, July 28 to August 10, 1928, showing the shortness of the period of effectiveness due to rapid evaporation and loss of the attracting substance; E, experiment 5, petroleum ether extract of 10 tips exposed by bottle method, August 1 to 16, 1928. Compare this with the same solvent by the can method in preceding experiment. The effective period was slightly longer and good catches were made, first, during the evaporation of the solvent, and then later by the residue. F, Experiment 6, anhydrous ether extract of 15 tips, July 28 to August 8; compare with petroleum ether extract in experiment 4

The area where traps were put out at Saugus, Mass., to check laboratory experiments, was a rolling wooded section of about 2 square miles, covered mostly with oak, averaging about 35 feet in height. Care was taken to place the cages or traps in the most lightly infested portion of this area in order to more nearly approach conditions prevailing in very lightly infested areas where it was expected the results might be applied. The traps were spaced approximately 150 to 200 feet apart. Table 8 gives the average yearly catch of male moths at the check traps, which contained at all times from 3 to 5 living, virgin females. As the females live only a few days, it was necessary to use from 30 to 45 living females per season for each of these traps. The trees bearing the traps were numbered (fig. 3, B) and carefully inspected to see that no natural infestation of pupae or female moths were near enough to affect the results. The results from these check traps are for comparison with the results from the extract traps when put out under similar conditions, and give a general idea of the density of infestation from year to year.

The extracts were prepared by clipping off into the solvent the posterior one to three abdominal segments of the females by means of a pair of sharp scissors. The extracts (except those carried over in cold storage) were held for a few days before being put in the field to allow for as complete extraction of the attracting substance as possible. Those referred to in Table 7 were put out by the can method. (Fig. 3.)

In a number of experiments vaseline and paraffin oil were added to the extracts for the purpose of decreasing the rate of evaporation of the active substance. In addition, geraniol, tincture of musk, a mixture of vegetable oils, soybean oil, and tincture of civet [18] were tried without the addition of female parts. Tincture of civet was also added in small quantity to benzene extract.

The catch of male moths by the various extracts increased, but not proportionally, as the number of female tips used in the solvent increased.

Of the various solvents represented in Table 7, xylene and benzene gave best results. The active substance was apparently more stable in these two solvents, since their extracts gave results that were more uniform, and attracted males over a longer period of time than did the extracts prepared from other solvents. Until 1927 one-eighth to one-half ounce of solvent was used for each experiment, but in 1927 1 ounce of solvent was used in most of the tests, and better results were secured. In 1927 the two experiments with 18 abdominal tips (fresh material) in 1 ounce of benzene caught an average of 495 males; one trap of benzene extract of 30-tip strength (1 year old) caught 442 males; and one of benzene extract of 30-tip strength (2 years old) caught 735 males. Two traps containing xylene and 30 abdominal tips (1-year-old material) caught an average of 323 males, while nine traps with fresh material of 16-tip strength under the same conditions caught an average of 242 males. The results indicated that benzene was better than xylene for preserving the material over a long period of time. That extracts of female tips deteriorate with age is clearly shown by individual experiments covering a

[18] Civet is a compound taken from the scent glands of the African civet cat.

period of years. In all cases it has been necessary to use more of the old extract to get results comparable to those given by the fresh material. The active substance has been preserved longer (seven years) in alcohol than in any other solvent. In most solvents it has not been active after the third year. It is very desirable that the active substance remain stable in storage for at least one year, since a considerable part of the extract prepared each year is being used in localities where the season is more advanced than in those from which it is necessary to obtain the female moths.

Alcohol extract gave slightly better results than the other extracts, but in either case it was necessary to use extract from a large number of abdominal tips to get a good catch of males. The results with carbon tetrachloride and chloroform were not very promising. Extracts containing one-fourth ounce each of paraffin oil, pine oil, geraniol, and vaseline gave poor results. Apparently, heavy oils retard the rate of evaporation of the attracting substance. Tincture of musk failed to catch any moths. Certain commercially prepared vegetable oils and extracts of beef fat failed to catch any moths when used in or without a solvent. A fair catch was obtained for a few days when an alcohol extract of female tips was poured on the bark of a tree.

The results with tincture of civet are very strange. When 10 drops were added to 1 ounce of xylene containing 15 abdominal tips only 15 males were caught; when 10 drops were added to 1 ounce of benzene containing 15 tips, the trap caught 611 males, next to the highest catch at any trap in 1927. When 2 drops were applied to the sticky material no males were caught; but when 2 drops were applied to cotton, 6 males were caught. When 1 c c was applied on cotton, 103 males were caught, most of which were attracted on the first day, and only 1 after the thirteenth day. In such heavy infestations an accidental catch of from 1 to 6 males sometimes occurs, allowance for which must be made in studying the results.

EXPERIMENTS IN 1928

In 1928, males were present in the field from July 20 to September 1 and were most abundant on August 2, which is about the time most of the extract traps were put out. During the season 264 traps were put out at Saugus, 161 of which were tests of extracts. The extracts were checked against one another, and against check traps containing at all times from 3 to 5 living virgin females.

Three methods of exposing the solvent and attractive substance for evaporation in the field were employed, namely, the can method previously described (fig. 3), the perforated-can method, and the bottle method. The perforated-can method resembled the can method in that the same can and cotton were used. It differed, however, in that the can was entirely closed, except for several holes punched in the bottom to permit the evaporation and escape of the solvent and attractant. The size and number of holes varied with the rate of evaporation of the solvent. The bottle method differed from the two methods just described in that the receptacle or container for the extract consisted of a 1-ounce or 2-ounce wide-mouthed bottle equipped with a cork stopper which held a glass tube in the shape of an inverted **V** with one end of the tube in the bottle and the other end outside and inclined toward the ground to prevent the entrance of rain. The odors passed out of the bottle through this tube. The tubes ranged in diameter from 1 to 30 mm, depending on the rate of evaporation of the solvent; the smaller tubes being used for the solvents which evaporated rapidly and vice versa.

The value of the different solvents varied considerably, the variation depending largely on (1) their ability to extract the attractive substance from the tips, (2) their effect on the stability of the attractive substance, and (3) their volatility. If pure fat solvents (as ether, gasoline, benzene, xylene) are used, most of the attractive substance is extracted from the tips in 1 day, and practically all of it in 5 days. In one instance where the tips were allowed to remain in considerable depth at the bottom of a small vial of xylene for 3 days without being mixed, the decanted solution caught about as many males as did the tips, which were covered and extracted with xylene again after the original solution had been decanted. In a number of other instances tips have been extracted twice both by ether and xylene, but the second extraction has caught only a small number of males, and this attraction may be due to the small quantity of the original liquid not removed from the tips by the filtering. Chemical tests have indicated that a small quantity of the attractive substance may be removed from the tips with 1 per cent hydrochloric acid, after the solvent has been filtered off. The 1928 material remained under liquid from 1 to 14 days, or an average of 5 days, before being put out. When an additional substance, as an oil, was added to a solvent containing tips, the addition was made after the tips had remained in the solvents for at least 1 day, so as not to interfere with the extraction of the attractant by the solvent.

The rate of evaporation of the solvents was determined as closely as possible at the laboratory by various methods of exposing known quantities to the open air for given periods during which the temperature, humidity, and amount of surface exposed were recorded. The rate of evaporation in almost all cases was most rapid at first, becoming less rapid as more of the solution evaporated. When the can method was used in the field the more volatile solvents, as ether, benzene, and gasoline, evaporated in from 10 to 36 hours, whereas some of the heavier oils, as cottonseed oil, required more than the length of one season for complete evaporation. Evaporation through the two other methods was much slower. By using the bottle method the evaporation of any of the solvents could be extended over the entire season. Evaporation by the perforated-can method was slower than by the can method, but was much quicker than by the bottle method. The bottle method proved to be the best method for ether, which was the most volatile of the solvents tried, but was inferior to the can method for all the other solvents, especially those which volatilized very slowly.

The experiments showed that the rate of volatilization of the solvent influenced the rate of volatilization of the attractant, e. g., when the solvent used was petroleum ether or ethyl ether, males were attracted for only about two weeks; but when benzene or xylene (which evaporates more slowly) was used as the solvent, males were attracted for as long as four weeks. (Fig. 4, experiments 3, 4, 5, and 6.) Although some of the attractant is given off during the evaporation of the solvent, most of it is given off from the residue after the solvent has evaporated. An example of this is shown graphically by experiments 3, 4, and 5. in Figure 4. One ounce of benzene was used with 30 tips in experiment 3, 1 ounce of petroleum ether with 15 tips in experiment 4, and 1 ounce of petroleum ether with 10 tips in experiment 5.

In experiment 5 the extract was evaporated by the bottle method and in experiments 3 and 4 by the can method. All of the solvent evaporated in less than one day in the case of experiments 4 and 6, and in about two days in the case of experiment 3, so at least 80 per cent of the total catch was made by the residue. In experiment 5 the rate of evaporation was reduced so that all of the solution was not evaporated until the sixth day, the catch after the sixth day being made by the residue. By reducing the rate of evaporation in experiment 5 the catch was nearly equal to those in experiments 4 and 6, and the period of catch slightly extended, although only 10 tips were used in experiment 5, as compared to 15 in experiment 4. The slow rate of volatilization of the attractant makes necessary the exposing of considerable surface in order to get a good catch of males. It is also quite evident from the daily-catch charts for 1927 and 1928 that when more than a few drops of oil was added to the solvent the rate of evaporation of the attractant was so retarded as to greatly reduce the efficiency of the extract. These observations support the view that the attractant is given off over a long period in the field by the hydrolyzing agent of the air. Among compounds that can be hydrolized are esters, proteins, and fats.

It is possible that over half of the attractant is lost from the extract while the males are not flying, since under most optimum weather conditions they are active for only about 9 hours out of 24 (9 a. m. to 6 p. m.). The most efficient means of liberating the attracting substance, if such were possible, would be to have all of it given off only during the time the males were active.

The promising results obtained in 1927 with tincture of civet, with and without the addition of female tips, suggested the use of a quantity of this material in 1928. Accordingly, civet, tincture of civet, and a synthesized product similar to civet were tried. As the chemical formula of this last (according to the chemist through whom this material was obtained) was not unlike that of oleic acid, many oils of this nature were tested. It was hoped that some substance might be found near enough like the attracting substance to either attract males without the addition of tips or increase the attraction when added to extracts containing tips. However, in all of the tests with civet and other compounds tested without the addition of female abdominal tips, negative results were obtained.[19]

A summary of the results from the more important experiments is as follows: Four traps containing 35 tips each of 1-year-old material in xylene caught an average of 194.5 males, and five traps of fresh

[19] The following substances, which gave poor to negative results, were exposed by themselves and in mixtures with various other substances both with and without the addition of tips: Tincture of civet, civet, oleic acid, cottonseed oil, amyl acetate, banana oil, corn oil, ethylamine, methylamine, benzonitril, quinoline, pine oil, oil of citronella, catnip oil, acetone, cheap gasoline, kerosene, cows' cream and yeast, honey, egg albumen, uric acid, and a mixture of all the materials listed above. A small quantity of hydrochloric acid, ammonium hydrate, water, and calcium carbonate were added separately to various substances.

material containing 15 tips caught an average of 361.5 males. Four traps with 30 tips of 3-year-old material in benzene caught an average of 140 males, and four traps of the same proportions of 1-year-old material caught an average of 224 males. These 17 traps were put out in an infestation known to be slightly heavier than that used for the other tests. The solvent which gave best results for fresh material in 1928 was a mixture of one-fourth ounce butyl acetate and three-fourths ounce of xylene, which caught 361 males, as against 350 caught by the check trap for that vicinity. High-test gasoline was the other outstanding solvent when 15 tips were used in 1 ounce of liquid. One trap of this strength caught 329 and the other 306 males in 34 days. Apparently 15 tips of fresh material, or 25 to 30 tips of 1-year-old material, per trap, is economically the best number for use in light or doubtful infestations. Three-fourths to 1 ounce of solvent per trap has given better results generally than other quantities.

EXPERIMENTS IN 1929

In 1929 the traps were put out by the can method, except where otherwise indicated, in a very lightly infested area at Saugus. Fifteen female abdominal tips were used in each case. Two traps with one-fourth ounce of butyl acetate in three-fourths ounce of xylene caught 149 and 204 males, averaging 176.5. When other proportions were tried the efficiency was reduced, as in 1928. Three traps with 1 ounce each of high-test gasoline caught 94, 190, and 203 males respectively, averaging 162.3 males per trap, whereas one trap with 2 ounces caught only 23 males. Two traps with a "special" gasoline caught 85 and 237 males, respectively, averaging 161. The gasoline was cheaper than the other solvents. Two traps with 2 and 1 ounces of benzene caught 122 and 182 males, respectively, averaging 152, and five xylene traps with 1 ounce of xylene each caught an average of 103 males. As the season began early and ended early not many moths were caught after August 15. Table 9 gives the more important results of the 1929 extract tests.

TABLE 9.—*More important results of experiments with attractants for the male gipsy moth, Saugus, Mass., 1929*

Solvent media (kind and quantity)	Date put in field	Days in field	Males caught
		Number	*Number*
High-test gasoline, 2 ounces [1]	July 19	37	23
High-test gasoline, 1 ounce	do	37	94
	July 22	35	190
	do	35	203
Average			162.3
High-test gasoline, 1 ounce (bottle method)	July 24	33	26
A special motor gasoline, 1 ounce	Aug. 10	15	237
Special gasoline, 1 ounce	July 20	35	85
A slow-burning gasoline, 1 ounce	do	35	43
	Aug. 10	15	8
Xylene, 1 ounce	Aug. 20	37	30
	July 19	37	87
	July 24	33	99
	July 23	34	138
	July 29	27	161
Average			103

[1] 15 tips or parts of abdomens of females used for each test except when stated otherwise.

TABLE 9.—*More important results of experiments with attractants for the male gipsy moth, Saugus, Mass., 1929*—Continued

Solvent media (kind and quantity)	Date put in field	Days in field	Males caught
		Number	*Number*
Xylene, 1 ounce, last segment of abdomen	July 22	35	62
Xylene, 1 ounce, last two segments of abdomen	do	35	115
Xylene, 1 ounce, whole abdomen	July 24	33	194
Xylene, 1 ounce, second and third segments from posterior end	do	33	10
Benzene, 2 ounces	July 19	37	122
Benzene, 1 ounce	do	37	182
Benzene, 1 ounce (bottle method)	July 24	33	9
Butyl acetate, one-fourth ounce, xylene, three-fourths ounce	do	33	204
	do	33	149
Butyl acetate, one-half ounce, xylene, one and one-half ounces	Aug. 7	18	90
Butyl acetate, one-eighth ounce, xylene, seven-eighths ounce	July 21	33	76
Butyl acetate, one-fourth ounce, benzene, three-fourths ounce	do	33	41
Butyl acetate, one-fourth ounce, high-test gasoline, three-fourths ounce	do	33	66
Amyl acetate, one-fourth ounce, xylene, three-fourths ounce	do	33	75
Quinoline, 5 c c (bottle method) [1]	do	33	0
Quinoline, 5 c c (can method) [1]	do	33	0
Civet, 1 g, on cotton [1]	do	33	0
Tincture of civet, 1 ounce [1]	do	33	0
Tincture of civet, 1 ounce (1-year-old material)	July 12	54	15
Check (average of three traps containing 34 living virgin females)			318

[1] Material in trap contained no tips.

Twenty-one traps were put out in 1929 with material that had been held in storage at an average temperature of about 2° C. for one year. None of the traps made a good catch, and only one caught a fair number (105) of males. The liquid portions consisted of xylene, benzene, gasoline, alcohol, petroleum ether, anhydrous ether, acetone, pine oil, catnip oil, oil of citronella, and tincture of civet, and contained from 15 to 30 abdominal tips per ounce. The material had been put in quart jars, the tops fastened down on rubber gaskets by clamps, and the jars kept in cold storage. A considerable proportion of the rubbers were affected by the solvents, and the evaporation from the different jars ranged from 0 to 60 per cent. The only liquids that did not undergo noticeable evaporation were pine oil, catnip oil, and oil of citronella. It seems likely that much of the attractive substance escaped from the jars in the same way as did the solvents, hence reducing the efficiency of the material. Capping the containers with cork stoppers wrapped with tin foil and sealed over the top with gelatine caps proved to be the best apparent remedy for preventing evaporation and leakage of gas or attractive substance during storage.

An attempt was also made to reduce the possible loss due to deterioration in storage by decanting the liquid from the tips and storing it separately until ready for use in the field. When the tips are left in the solvent it is possible that deterioration may result from such impurities as hair, excreta, fat, eggs, and chitin. When fresh extract containing rubber stoppers or iron rust was put out the attractant was apparently affected, as very few males were caught.

RESULTS WITH VARIOUS ABSORBENT MATERIALS

Six experiments were conducted in 1929 to obtain more data on the value of different absorbents in holding and releasing the active substance. Good absorbents should cause as little deterioration as possible. The can method with 15 abdominal tips was used in each case. Cotton was used in two of the traps, with 1 ounce of xylene as the solvent. In one case the cotton was packed tight in the can and in 37 days 73 moths were caught, whereas 187 moths were caught by the other trap with the cotton loosely packed. (Traps 41 and 42, Table 10.)

Traps 76 and 77 contained, respectively, 1 ounce of benzene and 1 ounce of gasoline extract poured over washed wool, but most of the solution dripped to the ground and only 55 and 30 moths, respectively, were caught in 30 days. In trap 82 only one-half ounce of gasoline was tried, and the wool allowed to soak in it overnight so that no loss by dripping occurred. This trap was put out August 9, after the period of maximum abundance of the moths. It remained in the field 20 days and caught 189 males, which is a favorable record under adverse conditions.

Kieselguhr or infusorial earth, which is an inert powder with good qualities as an absorbent and filter, was tried with 1 ounce of gasoline extract. The kieselguhr was held in place in the inverted can by a piece of cheesecloth stretched across the mouth of the can. (Trap 83.) Like trap 82, this trap was put out (August 7) after the maximum abundance of the moths had passed. The trap was in the field 22 days and caught 110 males. All except two of these moths were caught in the first four days. As rain wet the material through the cheesecloth within a few days after the experiment was begun, it is possible that it cut down the period of catch and incidentally the total catch. The results of these experiments are not final, but they indicate possibilities of finding a good absorbent by testing the value of various kinds and quantities of absorbent material. The results are given in Table 10.

TABLE 10.—*Tests of absorbent materials for holding extracts attractive to the male gipsy moth, 1929*

Absorbent used	Solvent media (kind and quantity)	Trap No.	Days extracted	Date put in field	Days in field	Remarks	Males caught
			Number		*Number*		*Number*
Cotton.	Xylene, 1 ounce	41	8	July 24	37	Packed tightly in can.	73
	Xylene, 1 ounce	42	8	...do...	37	Packed loosely in can.	187
Washed wool.	Benzene, 1 ounce	76	3	July 31	30	Nearly all of solution dripped to the ground.	55
	High-test gasoline, 1 ounce.	77	3	...do...	30	...do...	30
	High-test gasoline, ½ ounce.	82	6	Aug. 9	20	Extract poured in can, covered and allowed to soak for one day before putting out. Put out late.	189
Kieselguhr held in can by cheesecloth.	High-test gasoline, 1 ounce.	83	4	Aug. 7	22	Caught 108 moths in four days. Put out late. Very few moths flying.	110

TEST OF AGE OF FEMALES

Eight traps put out in 1929 with extracts from females of various ages yielded results that were interesting though subject to some uncertainty owing to the small number of tests. Experiments were conducted with abdominal tips clippd when the females were 1, 3, and 6 days old, one trap with tips from dead females 10 days old, and one with tips from female pupae. The tips were left in the solvents from four to seven days. No males were caught when tips from pupae or from females that had died naturally from old age were used. Of the other tests, extracts prepared from females 1 day old caught an average of 49 males, that from 3-day-old moths caught an average of 143.5 males, that from 6-day-old moths caught an average of 230.5 males. These results indicate that increased age, up to a certain point, greatly increased the amount of the attractive substance stored up in the females. The gasoline extract caught slightly more moths in each of the three instances than did the xylene extract. The three traps containing xylene caught a total of 403 males during the season, while the same number of traps containing high-test gasoline extract caught 443 males in a period several days shorter. The results are given in Table 11.

In 1931, 38 traps were put out with extracts from females that had just emerged. The results from these traps were compared with those from more than 38 traps containing extracts from females 2 to 6 days old. In all cases extract from females 1 to 4 days old caught at least 10 times as many males as extract from females that had just emerged when their tips were removed.

TABLE 11.—*Tests of extract prepared from abdominal tips of females of various ages, 1929*

Solvent media [1]	Date put in field	Days in field	Age of females	Males caught
		Number	*Days*	*Number*
Xylene	July 22	42	10 (dead)	0
Do.	do	42	[2]	0
Do.	do	40	1	48
High-test gasoline	July 29	33	1	50
Xylene	July 22	40	3	139
High-test gasoline	July 29	33	3	148
Xylene	do	33	6	216
High-test gasoline	do	33	6	245

[1] 15 female tips in 1 ounce of solvent used in each of these tests.
[2] Pupae were used in this test.

In 1930 several traps were put out with extract from females that had just emerged. A few of these traps caught no males, and none of the others caught more than a few. Several other experiments to determine the effect of age of virgin females indicated that the amount of attractant in the females increased for at least one day. It was found best not to keep them more than four days, as after that they began to die.

It is interesting that females seldom, if ever, mate twice or attract males after once mating, whereas virgin females afford strong attraction for males as long as they live. Twenty females were caged and records made of the daily catch and time of death. The last female to die lived 17 days and attracted males until death. A trap with extract from females that had just mated caught 129 males, whereas a trap of similar extract from females that had not mated caught 233 males. A trap with extract from females that had mated and laid eggs two days prior to the removal of their tips caught only 30 males, but extract from females that had mated from four to seven days previous to the removal of their tips caught no males. Extract from virgin females, two to six days old, caught many males, and an extract containing 15 female tips and 15 male tips caught 239 males, whereas an extract containing 15 male tips alone caught none.

RESULTS OF OTHER TESTS AT SAUGUS, MASS.

In 1930, 25 traps of 1929 material that had been held in cold storage for one year, and 97 traps of fresh 1930 material were put out. These traps caught a total of 11,624 males. The results from the more important solvents held in storage are given in Table 12. Observations indicated that extracts of tips of females that were less than 12 hours old did not give good results, but one trap containing only 15 tips in xylene from females approximately 24 hours old caught 214 males, which is about equal to the catch with fresh (1930) material. Six traps containing tips from females less than 12 hours old in high-test gasoline caught an average of only 8 males. It is not known whether this result was due to the females not being old enough at the time the tips were removed or to a lack of stability of the attractant in this solvent under the cold-storage conditions. The storage containers were sealed with cork stoppers wrapped in tin foil and covered with gelatin caps, and no noticeable loss of extract by evaporation could be observed.

TABLE 12.—*Tests of attractant material that had been held in cold storage for one year, Saugus, Mass., 1930*

Solvent	Female tips per trap	Traps	Age of females	Average catch
	Number	*Number*		*Number*
High-test gasoline	15	3	Less than 12 hours	6
	30	3	do	10
Xylene	30	4	do	44
	15	1	Less than 24 hours	214
Benzene	15	2	Less than 12 hours	73
	30	1	Less than 24 hours	173
	15	1	do	187
Butyl acetate one-fourth ounce, xylene three-fourths ounce.	15	1	24 to 48 hours	144

In the tests made at the same time with the fresh material the females from which the tips were obtained were at least one day old but not over four days old. Each trap contained 15 tips in 1 ounce of solvent, and all were put out under practically identical conditions.

The results of these tests are as follows: Two traps with gasoline from the Texas oil fields as a solvent caught an average of 292 males; two traps with a special motor gasoline caught an average of 254 males; and six traps with a high-test airplane gasoline [20] caught an average of 240 males. One trap with toluene as

[20] As the distillation points of lots of the same brand of gasoline are apt to vary, it has been the practice during the last two years to purchase two or three 25-gallon samples from different lots, and any one of these proving to be a good solvent will furnish a supply sufficient for several years.

the solvent caught 245 males; eight traps with xylene as the solvent caught an average of 196 males; and two traps with benzene as the solvent caught an average of 152 males. Two check traps, with approximately 30 living females each, caught an average of 494.5 males.

In one experiment the genitalia and tissue immediately surrounding the opening of the copulatory pouch and oviduct of 15 females was put out in one trap, and the last three abdominal segments of the same females without the tissue used in the trap just mentioned put out in another trap. The former trap caught 279 males and the latter 20.

As in previous years the traps of 1931 were spaced 100 to 200 feet apart along trails in a forest area of approximately 600 acres. One hundred and fifty-one traps were put out, or about one trap to 4 acres. A total of 7,833 males were caught, an average of 51.87 per trap, or about 13 per acre. The infestation was extremely light, for only two egg clusters were seen during the daily visits to the traps, and two men collecting for three hours were able to find only 12 pupae (11 male and 1 female). The fact that more male than female gipsy moths reach the adult stage in the field makes it easier to locate infestations by trapping than if the reverse were true.

In 1931 a double-funnel or double cone-shaped trap was devised. It turns easily on an axis in such a way as to hold the point of one cone toward the wind, and the other cone in the opposite direction. About 90 per cent of the males trapped were caught by the cone with the point toward the wind.

OBTAINING FEMALE ABDOMINAL TIP MATERIAL

The development of methods that will permit the coverage of a greater area with traps, and at a lower cost, is an important research problem. There are two methods of attack: One is improving the quality of the extracts and methods of exposing in the field so that the traps will be more dependable and so that a given amount of material can be made to cover a greater area. This requires the discovery and use of the best solvents, clipping the tips when the females reach the age at which they yield the greatest quantity of attractive substance, prevention of evaporation and deterioration in storage, and the development of better traps. The other is obtaining a larger number of female moths, which may be accomplished by either one or by both of two ways, i. e., by improving collecting methods and increasing the collecting force, or by increasing the percentage of emergence from the pupae collected. Table 13 shows that 945,314 pupae collected yielded only 148,965 moths, which is an issuance of 15.8 per cent, or about 1 moth for each 6 pupae. This mortality of 84.2 per cent was due to disease, parasites, and to unknown causes. Considerable loss results from handling large amounts of material. If by improved handling methods the ratio of issuance could be increased to 2 moths for each 6 pupae, instead of 1 out of 6, it is at once obvious that the area to be covered could be doubled. During some years the pupae were collected in small shallow boxes and transported to a central collecting station where they were spread out in thin layers on the floor (fig. 5) or in trays (fig. 6), and the female abdominal tips clipped daily as the moths issued. The amount of available material varies from year to year owing to natural control factors and facilities for collecting.

TABLE 13.—*Data on collection of female pupae and the number of moths obtained*

Year [1]	Pupae collected	Moths issuing		Remarks
	Number	*Number*	*Per cent*	
1920	340,800	30,929	9.1	Collected by 8 men working 19 days.
1923	225,075	26,500	11.8	Collected by 8 men in 21 days.
1925	70,439	24,560	30.9	
1928	300,000	66,976	22.3	Collected by 8 men in 21 days.
Total	945,314	148,965	15.8	

[1] In 1924 a total of 34,860 moths were obtained; but as the exact number of pupae from which these issued was not recorded, they have not been included in the tabulations.

In 1929 a small box, with two sliding drawers that were divided into many small partitions in which the pupae were placed, was used for collecting. An increased percentage of emergence was obtained,

FIGURE 5.—Female gipsy-moth pupae spread out on canvas. As the moths issued and crawled up the canvas sides they were picked off and the tips of their abdomens removed

but this value was offset by the fact that the rate of collecting was reduced. In 1931 the method of collecting and rearing pupae was greatly improved, and many more moths were obtained then than in previous years. The pupae were carefully removed from the trees and placed between thin layers of cotton batting in the trays of collecting boxes, and carried to a ventilated room of near-cellar temperature ranging from 60° to 75° F., close to the collecting point, where they were transfered in the cotton layers to large, coarse, wire screened trays supported by large racks. By this method of handling, the pupae were allowed to become entangled in and attached to the

FIGURE 6.—Female gipsy moth pupae spread out in coarse wire screened trays. The moths are removed and counted soon after emerging. The two boxes at the bottom of the rack contain moist earth into which larvae of the parasite *Sturmia nidicola* Towns. fall and are saved and later returned to the field.

cotton fibers in much the same manner as they were found naturally on the tree. The layers of cotton also helped to prevent injury by crushing. By having the rearing place at or near (within 5 miles of) the point of collection, injury by transporting bulk collections long distances (as 60 miles or more) was reduced to a minimum, and the cool room used for rearing prevented loss by hot, dry air. The females were counted and held for one to two days before their tips were removed.

Other collections of pupae were made as follows, and transported for about 60 miles by truck to the Melrose Highlands, Mass., laboratory and placed in a shaded screened cage: 2,173 on cotton layers as described above, 5,560 on sawdust, 3,800 on cotton in a partitioned box similar to an egg crate, and 1,550 on wool layers. Because of improved methods of handling, a good issuance was obtained in all cases. The percentages of pupae giving moths were as follows: On sawdust 38, on cotton 42, on wool 50, and on cotton in the partitioned crate 52. Only 11 per cent of the pupae had become moths in 1930, owing to a greater mortality from parasites and disease than in 1931, and to imperfections in the handling methods which were eliminated in 1931.

One thousand nearly mature female larvae were collected in 1931 and fed oak leaves until they pupated. Although considerable wilt disease developed, an issuance of 19 per cent was obtained.

The following figures will show the quantity of material needed to cover a given area in case it were advisable to use attractants on a large scale: In 1928 eight men collected 300,000 female pupae in 21 days, from which 66,976 moths issued as available abdominal-tip material. This made enough extract for 2,233 traps at 30 tips per trap, or 4,465 traps at 15 tips per trap. As the average town has about 70 miles of road, this amount of extract would cover 8 towns, if "30-tip" strength were used in traps one-fourth mile apart. If "15-tip" strength were employed, 16 towns could be covered, and still larger areas by putting the traps farther apart. In all towns, however, there are large areas with little or no tree growth so that the possible infested area that could be covered is greater than is represented by the above figures.

USE OF EXTRACTS TO LOCATE NEW INFESTATIONS IN NEW ENGLAND

A varying number of extract traps were put out in New England at or near the quarantine line each year from 1923 to 1929. Those put out in 1923, 1924, and 1925 gave very poor results. In Essex, Vt., only one male was attracted in 1924, and in Sheffield, Mass., only one in 1925. In a few cases infestations existed within one-half mile of traps that attracted no males. A small quantity of vaseline was added to the benzene extract in 1925 to prevent the solution from evaporating so rapidly, but this proved detrimental. Better results were secured as improvements of the extracts were made, and in 1926 a large number of traps were put out. The numbers of males caught are shown in Table 14.

TABLE 14.—*Summary of catches at extract traps at or near the New England quarantine line, 1926*

Location	Egg masses found		Distance to nearest traps	Solvent	Males caught
	Old	New			
	Number	*Number*	*Miles*		*Number*
Norfolk, Conn.	3	1	½	Xylene	0
			¾	do	1
			¾	Benzene	1
North Canaan, Conn.	3	29	½	Xylene	0
			½	do	0
			½	Benzene	0
	1	1	½	do	0
			½	do	0
Sheffield, Mass.	10	26	½	do	0
			¾	do	0
	6	7	⅝	do	0
			¾	do	1
			⅛	Xylene	1
			½	do	0
Sandisfield, Mass.	40	26	⅝	do	1
			¾	do	0
			¾	do	0
			¼	do	0
			¼	do	3
Barkhamsted, Conn.	5	55	½	do	4
			½	do	5
	5	8	⅝		
Canton, Conn.	1	2	½		10
		473	¾		
	----	3	⅜	Xylene	8
	39	30	⅜		
	22	140	⅜	do	8
Granby, Conn.	12	29	¼	do	10
	199	990	⅜	do	16
	425	495	½	do	10
	2	1	½	do	0
		1	1⅛	do	1
			5/16	do	1
Colebrook, Conn.	8	1	⅝	do	0
			½	do	0
Simsbury, Conn.	3	12	⅝		1
			⅞		0
	12	86	1/16	Benzene	1
	37	538	¼	do	1
			¼	do	1
			⅝	do	1
	12	24	⅛	do	0
Essex, Vt.			¼	do	0
			⅛	do	6
	4	24	1/10	do	0
	22	76	½	do	2
			7/16	do	0
	5	43	1/16	do	0

Table 14 shows that both positive and negative results were secured. Twenty-three traps that were within 1 mile of infestations attracted 1 or more males, whereas 21 traps within similar distance from infestations did not catch males. At many traps catches of from 1 to 16 males were made, and subsequent scouting revealed infestations of one or more newly deposited egg masses. The larger catches of 8, 10, and 16 males, respectively, per trap were made in Granby and Canton, Conn., where the infestations were one-half mile, or less, distant. The cages were not attended daily, and the males were removed at the end of the season; consequently no relation could be observed between the catches and the prevailing winds to determine why several of them did not attract. It is possible that alternate-year changing of the trap locations in each vicinity might take care of much of this variation.

USE OF EXTRACTS IN NEW JERSEY

In 1920, when the gipsy moth was first found in New Jersey, 21 traps containing extract of abdominal tips of females were put out near the center of the original colony and a few miles distant. This was done to ascertain whether this method would be of help in locating new colonies.

Material preserved in 1917 and 1919 in 95 per cent alcohol or in petroleum ether was used. Strong dosages were used, ranging from 2 to 4 teaspoonfuls of extract, representing from 215 to 479 abdominal tips, which proved to be more than was necessary. A total of 85 males were caught in 14 of the 21 traps. Two of the traps containing alcoholic extract attracted 18 and 23 males, respectively. These held a higher concentration, however, than the contents of those of petroleum ether, the best of which attracted seven males. Subsequent scouting showed that there were scattered gipsy-moth colonies throughout the section where the traps were put out, and in some cases a colony within a few hundred feet of the trap.

The object of the campaign against the gipsy moth in New Jersey was to exterminate the insect. The material used in these traps was necessarily collected and preserved in New England the summer previous to its use in New Jersey, as the season is earlier in New Jersey. The abdominal tips were later preserved in gasoline, xylene, and benzene, which were found to be the better solvents. The 1-year-old material was used at the rate of 30 to 40 female abdominal tips per trap.

The field scouting in New Jersey was very carefully done each year, and the scouting in 1924–25 resulted in the finding of some colonies in the vicinity of traps that did not attract any males in the summer of 1924. The following statements give the results of the scouting about these traps and the solvents used:

Hillsboro. Infestation of 10 new egg masses found in Franklin Township, five-eighths mile from cage containing benzene and crushed tips.

South Brunswick Township. Infestation of 35 egg masses found one-eighth mile from cage containing gasoline and uncrushed tips. Infestation of three new egg masses found five-eighths mile from cage with benzene and crushed tips.

Bridgewater Township. Infestation of one new egg mass found seven-sixteenths mile from cage with benzene and uncrushed tips. Another cage containing gasoline and crushed tips was also located seven-sixteenths mile from the above infestation. Infestation of one egg mass found 100 and 150 feet from two cages containing gasoline and crushed tips.

The results of the above attempts were all negative, and it is impossible to explain this satisfactorily unless it was due to the deterioration and evaporation of the extract in storage and in transit. Improvement of extracts and methods in general have been made, and more encouraging results were obtained in New Jersey with the 1926 experiments, as is shown by Table 15.

Table 15.—*Results of experiments with benzene extracts to attract male gipsy moths in New Jersey, 1926*

Township	Egg masses found		Distance of nearest cage	Males caught
	Old	New		
	Number	*Number*	*Miles*	*Number*
Bridgewater	33	88	1¼	1
			⅛	2
	19	5	1	2
			1¼	1
	4	3	¾	1
Hillsboro	0	3	⁵⁄₁₆	1
Bernard	33	88	[1] ¾	1
Warren	4	7	1¼	1
	47	68	⅞	1
	1	4	[2] 1¹⁄₁₆	1
	12	10	([3])	2
Total	153	276		14

[1] Another nearer cage failed to attract.
[2] Two nearer cages failed to attract.
[3] At cage.

Table 15 shows that 14 males were caught. This information proved of considerable value to the eradication project, especially since three traps located in parts of the townships of Bridgewater, Bernard, and Warren, which recent records had indicated were free of the gipsy moth, attracted one male each. A catch of three males in a heavily wooded locality indicated the presence of one or more infestations in the neighborhood. Some special scouting was done, and a colony of 33 old and 88 new egg masses was found within 1¼ miles of the traps attracting. Instead of being left to further reproduce until plans for scouting materialized, the colony was immediately treated, and then sprayed in 1927. Other colonies were also found nearer than this to the attracting cages, as shown in the table. Some of the traps did not catch any males, and no colonies were found in these localities. Since 1927 very few colonies have been discovered in New Jersey, and in 1930 and 1931 none were found.

SUMMARY

Male gipsy moths are strongly attracted to living virgin females. A scent is given off from the vicinity of the copulatory-pouch opening and is sensed by the male through its antennae.

Some males fly as far as 2.38 miles, but usually the distance of flight is much shorter; and catches of males are seldom made at traps over one-half mile distant from a colony. The percentage of released males recovered is dependent upon a number of factors, such as distance, wind, temperature, and topography.

Certain extracts made from the region of the female genitalia attract males, for the attractant is soluble in a number of fat solvents, as ether, benzene, xylene, and gasoline. None of the attractant was found in glands of the reproductive system internally, although in the vicinity of the copulatory-pouch opening setae and pores peculiar to this region were found, and it is possible that the attractant arises here, for extracts of this portion of body-wall tissue attracted many males. The attractant is of a complex fatty nature,

and is saturated. When exposed, it is active for several weeks, a period equivalent to the flight season of the males—owing to the continuous generation of the attractive compound by hydrolysis. It is fairly stable under present storage methods, and from two-fifths to three-fifths of its value is retained at the end of one year.

At the moment the female emerges little attractant can be extracted, but the quantity rapidly increases thereafter for at least one day. Virgin females attract males as long as they live, but females that have mated generally do not attract them. Although considerable of the attractant may be extracted from females soon after they have mated, the quantity rapidly decreases.

The percentage of female moths obtained from bulk pupal collections was greatly increased by placing the pupae between thin layers of cotton or wool batting in partitioned collecting boxes, and then transferring them to a ventilated room of moist air at a temperature of 60° to 75° F. The room should be located near the point of collection to prevent injury in transporting them long distances. The best results were obtained when the moths were allowed to age for two days before the tips were removed.

Results indicate that the practical value of extract traps may be improved further by (1) increasing the number and proportion of female moths obtained from available pupae, (2) removing the female abdominal tips at the age when they yield the greatest quantity of the attractive substance, (3) improving the extracts, (4) preventing deterioration in storage, and (5) determining the best method and best absorbent material for exposing the extracts in the field.

The use of an extract in the traps instead of living females has the following advantages: (1) The extract can be preserved for one or more years and be available for use in newly infested localities where the season is earlier than in the place from which living females would have to be obtained, (2) it is not necessary to visit the traps except to put them out and remove them at the end of the season, and (3) the danger of starting new infestations is eliminated.

Some new and important infestations in the border of the infested territory and in New Jersey were discovered by the use of traps containing extracts of female abdominal tips.

ORGANIZATION OF THE UNITED STATES DEPARTMENT OF AGRICULTURE WHEN THIS PUBLICATION WAS LAST PRINTED

Secretary of Agriculture	ARTHUR M. HYDE.
Assistant Secretary	R. W. DUNLAP.
Director of Scientific Work	A. F. WOODS.
Director of Regulatory Work	WALTER G. CAMPBELL.
Director of Extension Work	C. W. WARBURTON.
Director of Personnel and Business Administration.	W. W. STOCKBERGER.
Director of Information	M. S. EISENHOWER.
Solicitor	E. L. MARSHALL.
Bureau of Agricultural Economics	NILS A. OLSEN, *Chief.*
Bureau of Agricultural Engineering	S. H. MCCRORY, *Chief.*
Bureau of Animal Industry	JOHN R. MOHLER, *Chief.*
Bureau of Biological Survey	PAUL G. REDINGTON, *Chief.*
Bureau of Chemistry and Soils	H. G. KNIGHT, *Chief.*
Office of Cooperative Extension Work	C. B. SMITH, *Chief.*
Bureau of Dairy Industry	O. E. REED, *Chief.*
Bureau of Entomology	C. L. MARLATT, *Chief.*
Office of Experiment Stations	JAMES T. JARDINE, *Chief.*
Food and Drug Administration	WALTER G. CAMPBELL, *Director of Regulatory Work, in Charge.*
Forest Service	R. Y. STUART, *Chief.*
Grain Futures Administration	J. W. T. DUVEL, *Chief*
Bureau of Home Economics	LOUISE STANLEY, *Chief.*
Library	CLARIBEL R. BARNETT, *Librarian.*
Bureau of Plant Industry	WILLIAM A. TAYLOR, *Chief.*
Bureau of Plant Quarantine	LEE A. STRONG, *Chief.*
Bureau of Public Roads	THOMAS H. MACDONALD, *Chief.*
Weather Bureau	CHARLES F. MARVIN, *Chief.*

This bulletin is a contribution from

Bureau of Entomology	C. L. MARLATT, *Chief.*
Division of Forest Insects	F. C. CRAIGHEAD, *Principal Entomologist, in Charge.*

U. S. GOVERNMENT PRINTING OFFICE: 1932

For sale by the Superintendent of Documents, Washington, D. C. - - - - - Price 5 cents

END