PAPERS AND PROCEEDINGS OF THE 1951 ANNUAL MEETING—Part

BIOLOGICAL EFFECTS OF DDT APPLICATIONS ON TIDAL SALT MARSHES

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Consequent to the increased use of DDT and other insecticides for the control of salt-marsh mosquitoes, there has been a growing concern over possible hazards to fish and wildlife. In an effort to evaluate properly any harmful features of such programs, and to seek means for minimizing any dangers to beneficial life, the Fish and Wildlife Service, in cooperation with other agencies, initiated an investigation in 1949. This report summarizes the results obtained during the first two years (1949-1050) of such a study.

Concerning salt-marsh mosquito control with DDT, little is known of the direct or indirect damage to fish, marsh birds and waterfowl, and to such commercially important invertebrates as blue crabs, clams, oysters, and bait shrimp. present study has aimed to determine the extent of damage sustained by wildlife with sprayings comparable to those applied for control of mosquito larvae. However, due to the small amount of population work on many of the organisms encountered, a considerable portion of the investigation necessarily was directed toward the development of suitable study methods.

THE AREA AND METHODS

Studies were conducted on the salt marshes of Brigantine National Wildlife Refuge, Oceanville, New Jersey. In addition to the Fish and Wildlife Service. several federal and state agencies participated. The Bureau of Entomology and Plant Quarantine of the U. S. Department of Agriculture furnished insecticide and the plane; the New Jersey Agricultural Experiment Station studied effects of the

aerial applications upon insect pests of the area and personnel of the New Jersey Fish and Game Division assisted materially in several phases of the project.

In 1949, four study areas were established and treated by plane at rates of 1.6, 0.8, 0.4 and 0.2 pounds of DDT per acre respectively. Two applications were made, the first between August 17 and 19 mostly at high tide and the second on September 15 and 16 at low tide. Each plot included 100 acres except for the 1.6 area which was 50 acres. Realignment of the plot boundaries before the second treatment resulted in some portions of each of the areas being treated only once and other parts twice. For comparison, studies were also made throughout the summer on the fluctuations of animal populations in suitable unsprayed check areas.

Since the 1949 studies clearly established the consequences to aquatic life of the 1.6 pounds per acre application, this dosage was omitted from the 1950 program. However, in this second year of study the former 0.8 pound per acre plot received an application of 1.0 pound per acre, and the 0.4 and 0.2 areas of 1949 were treated in 1950 at rates of 0.5 and 0.25 pound per acre respectively.1 Three sprayings were made on each area at low tide on July 6, August 4 and September 3. Although the 1.6 area of 1949 was not re-treated in 1950, follow-up studies were conducted to determine the presence of any continued effects.

¹ Hereafter these areas or dosages are referred to as the 1.6, 1.0, 0.8, 0.5, 0.4, 0.25, and 0.2 areas or dosages.

During the first year the spray used was Shell ACX-331 containing 10.75 per cent DDT in light fuel oil with an auxiliary solvent. Different dosages were obtained by varying the volume of spray applied. With this procedure, higher dosage plots received more oil solvent than the lower dosage areas. Since oil is toxic to certain forms of life, greater or lesser kills might be expected according to the amount applied. This fact recommended a change in the 1950 program, and during the second year, 20, 10 and 5 per cent oil sprays were made up for the 1.0, 0.5 and 0.25 areas respectively. Consequently, equal quantities of spray per acre were applied on each of the areas, the amount of DDT being the only variable.

Pre- and post-spray studies of populations and of animal behavior in sprayed and untreated areas were the chief investigative methods employed, and techniques used are described at appropriate places in the text. Since the majority of invertebrates are more readily affected by DDT than vertebrates, it was felt that spraying effects on the latter would be largely indirect and evidenced primarily through disturbances to invertebrate populations. Consequently, invertebrates received major emphasis in this study. Many of these serve directly or indirectly as a source of food for vertebrates, and a few, such as the blue crab, are of commercial value.

OBSERVATIONS ON BIRDS

Gulls.—Greatly increased numbers of gulls, mostly laughing gulls, Larus atricilla, were usually seen at low tide on the day following spraying. These increases were noted in areas where considerable mortality to fish, crabs and shrimp had occurred. The actions and cries of feeding gulls undoubtedly attracted birds from considerable distances as up to 500 were sometimes seen in an area. These concentrations largely disappeared by the second day following treatment. Thereafter, in some cases, a below-average number of gulls were present in the sprayed areas. Strip counts on the 1.6 and 0.8 plots before and after spraying in August showed an 85 per cent decrease in number of gulls following treatment. No harmful effects to these birds could be discerned, even though they consumed large quantities of DDT-contaminated animals. Usual numbers were again seen in the areas within several weeks following spraying.

Clapper Rail.—Though it was difficult to observe feeding habits of clapper rails, Rallus longirostris, it was evident that they consumed many DDT-affected fiddler crabs. This was indicated by the increased number of large claws of the male crabs following spraying. These are commonly discarded by rails when eating fiddlers. Surveys of the plots after treatment revealed no dead birds in 1949, but in 1950 a young rail about one week old was found dying in the 0.25 area, 40 hours after the August 4 spraying. Upon death, tissues from this specimen were analyzed and found to contain DDT in quantities sufficient to suggest acute poisoning. Since this bird was recovered on the lowest dosage plot, it seems reasonable to assume that other deaths may have occurred on areas of heavy dosage.

Following the July, 1950 application, observations were made on a total of 11 nests located in treated plots, five of which were in the 1.0 area. All nests produced successful hatches indicating no apparent effects to parents or young. Several rails collected at various times after treatment had well-filled gizzards, indicating that there was no shortage of food.

Herons.—Increased numbers of herons and egrets entered some of the study plots for a few days after spraying. None were observed eating dead fish, nor were there any indications of mortality to these birds.

Swallows.—Tree swallows, Iridoprocne bicolor, while very common before the August, 1949 spraying in the 1.6 and 0.8 areas, decreased greatly thereafter. Strip counts showed declines in these areas of 52 and 41 per cent respectively. Meanwhile, increased numbers of birds were observed feeding in nearby untreated areas. However, this was only a temporary change as the usual numbers were again seen over the sprayed plot before the next

treatment. Swallows were less numerous before spraying in the lower dosage areas and any changes thereafter were not noted.

Song Birds.—The effects of DDT on small resident song birds including the long-billed marsh wren, Telmatodytes palustris, sharp-tailed sparrow, Ammospiza caudacuta, and seaside sparrow, A. maritima, were determined by strip counts in the higher dosage areas. An average of 26 birds in the 1.6 area, and 29 birds in the 0.8 area were present before spraying. Afterwards averages of 26 and 32 birds respectively were seen, indicating no significant changes. No observable harm resulted to several nestlings and young birds just out of the nest in the high dosage areas. Collection and examination of both young and adult specimens five days after spraying showed that all had well-filled gizzards.

Effects on Fishes

Investigations were made of DDT effect upon the common fishes of the salt marsh, principally killifish. Two species were especially typical: Fundulus heteroclitus, found abundantly in all aquatic habitats, and Cyprinodon variegatus, which oc-

curred largely in ponds.

General observations indicated that killifish were readily affected by DDT spray, the results being largely in accord with the dosages applied. At rates of 0.8 pound per acre and above, poisoning symptoms were observed within four hours after low-tide treatments, and at lower dosages within nine hours after spraying. maximum number of dead and affected fish were usually seen 18 to 24 hours following spray application. In the low dosage areas, all visible effects of spraying had disappeared by the second to fourth day after treatment while in the 1.6 and 0.8 areas dead fish were seen until the fourth or fifth day.

Heavy losses of fish occurred as a result of the 1.6 dosage spraying. Small Fundulus less than 1½ inches long made up the majority of the kill in the creeks and ditches but there were a number of larger individuals up to 3 inches in length. Mor-

tality was not complete and many fish were seen during succeeding tides. In ponds the kill was greater, being nearly complete in the shallow bodies. This difference was probably associated with the greater concentration of spray in the shallow water areas and the greater possibility of contact by fish.

Lower mortalities occurred in the 0.8 area and the number of dead and affected fish was only about half of that present in the 1.6 area. Even less mortality occurred in the 1.0 area in 1950 and here the losses were only a little greater than those observed in low-dosage areas. though a number of small fish were affected within four to five hours following the 1.0 spraying, most of them recovered. The difference between the two years' spraying appears to be due to the greater volume of oil used in the 1949 operations. The effects of spraying at the lower dosages were much reduced and were confined to the smaller sized fish in the shallow ponds and ditches. A number of affected fish were noted in the areas but only a few succumbed. No mortality or evident harm to fish was observed in the check areas before or after sprayings.

Observations of effects of the various sprayings were confirmed, in a general way, by cage experiments although results from the latter were clouded by mortalities from handling and by probable weakening of fish through confinement. However, handling mortality was largely eliminated during the second and third treat-

ments in 1950.

These experiments showed about a 70 per cent loss of fish in both creek and pond cages at the 1.0 pound per acre application. In the 0.5 and 0.25 areas, small fish (under 2 inches in length) suffered about 50 per cent reductions in both creek and pond habitats. Losses to larger fish (over 2 inches in length) in the lower dosage areas were somewhat less. Creeks showed a 40 per cent mortality in the 0.5 area and half of that in the 0.25 area. In ponds, the greatest losses to large fish occurred in the 0.25 area where a 45 per cent mortality was observed as compared to a 20 per cent

loss in the 0.5 area. *Cyprinodon* killifish proved to be more resistant to DDT spray than *Fundulus* in the pond tests.

Cage studies were also made to determine if fish in the main drainage creeks outside each of the treated areas were affected by DDT spraying. No mortalities occurred among large or small fish placed at an average distance of about 1800 feet beyond the far ends of the plots.

A more natural means of confinement used in the 0.4 and 0.2 areas consisted in screening-off sections of creeks which retained considerable water at low tide. Dead and live fish subsequently recovered indicated a mortality of 86 per cent in the 0.4 area following spraying. However, almost no mortality was observed in a similar experiment in the 0.2 area.

STUDIES ON INVERTEBRATES

Insects.—Net sweepings along established courses, before and after DDT sprayings, gave indices of the populations of various kinds of insects in the marsh and also denoted the comparative decline in numbers following treatments. Of the insects present on the salt marsh, leaf-hoppers (Homoptera) and flies (Diptera) were found to be most abundant in numbers of individuals.

Severe reductions in populations resulted from DDT spraying. Declines following a single spraying ranged from about 15 to over 80 per cent, the losses being roughly proportional to the dosage applied. True bugs (Hemiptera) were almost completely eliminated. Leafhoppers and flies were more resistant, especially the former. Survival of various species proved to be distinctly greater in tall grass than in short grass, probably due to the greater screening effect of the higher vegetation.

The duration of spraying effects on insects was found to depend upon the dosage applied and the time of the year. Areas treated with high dosages (0.8 and above) in mid-August showed continuing declines in populations through September followed by complete recovery the next summer. However, the residual effects from treat-

ments in early July disappeared largely in less than a month although populations had still not attained pre-spray levels by the end of the summer. On the other hand, sprayings at lower dosages (0.5 and below) were followed by complete population recovery within seven weeks.

Repeated treatments strongly curbed increases in insect numbers. Populations in the 0.5 and 0.25 areas were reduced about 90 per cent by the end of the summer as compared to pre-spray populations while during the same period the check area populations rose approximately 80 per cent.

Blue crabs (Callinectes sapidus).—These animals proved to be very susceptible to DDT applications and in some cases suffered severe losses. In the watercourses, these reductions as well as other effects of DDT spraying were demonstrated by means of general observations, by trapping crabs in commercial type crab pots and by periodic confinement and counting of crabs in screened sections of creeks and ditches.

Screened channel sections were used to help appraise crab mortality in conjunction with the second 1949 spraying and all the 1950 treatments. Screen barriers extending above the high tide level were erected within the channels of creeks and ditches of each area and enclosed portions of individual creeks or of creek systems ranging up to 4300 feet in length. screens were adjustable and could be raised and lowered to permit the passage of crabs or to enable confining them at low tide for counting purposes. All enclosed creek channels contained sufficient water at low tide to maintain crabs. Dead individuals were removed as they were found.

Information on spraying effects on blue crabs was also obtained by means of crabpot catches taken at weekly intervals throughout the 1950 season. Resulting data, although not as accurate as those obtained from the enclosed creek systems, tend to confirm the findings obtained by other methods.

General observations together with screened-channel studies and crab-pot

catches showed that the greatest losses among crabs occurred at the higher rates of application (0.8 pound per acre and above). Low-tide sprayings with these dosages produced typical symptoms of DDT poisoning within two hours after However, the greatest effect from spraying at these and at lower dosages did not occur until 18-24 hours later. Surveys at this time in creeks and ditches of the 1.6 area revealed a very heavy mortality of crabs that had been present at the time of spraying. The few live crabs that were seen showed effects of DDT poisoning. Probably some affected crabs had left during preceding ebb tides but the numbers of dead crabs that were found indicated that the kill within the area had been nearly complete. Results in the 1.0 and 0.8 areas were similar although mortality was not as sudden and about one-third of the crabs present 24 hours after spraying were still alive. Crabs entering these areas during subsequent incoming tides suffered mortalities for a period of about two weeks.

Repopulation was slow and was approximately half completed in four weeks. Complete recovery in the 1.6 area treated in both August and September did not occur until the middle of the succeeding summer. The 0.8 plot of 1949 received a single 1.0 pound treatment again in July, 1950 and at the end of this second season its average population was approximately one-fifth as great as the population prior to treatment the previous year. Seven weeks were required for the population to rebuild to the 1950 pre-spray level.

Effects of treatments at the lower dosages were more variable. General observations during the first season indicated light mortality in the 0.2 area and only a moderate kill in the 0.4 area. However, confinement of crab populations in the creek channels of these areas revealed kills of 60 per cent in the 0.2 and 90 per cent in the 0.4 area during periods of one and two weeks respectively.

Channel enclosures operative during the period of a single tide showed losses of over 80 per cent in the 0.5 area and from 20 to over 40 per cent in the 0.25 area. DDT mortality in both areas was of about seven days' duration. The number of dead crabs recovered in the 0.5 area following spraying did not account for the total number missing and indicates that either crabs were dying outside the creek system or that they did not enter in the usual numbers. Within a day or so after the termination of mortality, crabs were as plentiful in the 0.25 area as before spraying. In the 0.5 area this process required two additional weeks.

Pond habitats showed greater losses than creeks and ditches. Nearly complete mortality was observed following single treatments of ponds at the high dosage rates. After the third spraying a survey of 19 ponds in the 0.5 area revealed no living crabs while at the same time only two could be found in 16 ponds in the 0.25 area. As in the case of fish, more severe kills in these low dosage areas were noted in shallow ponds than in deep ponds. The lack of flushing action, except by spring tides, undoubtedly accounted for the greater kills observed in ponds as compared to creeks and ditches. Tides, of course, also regulate the time and frequency with which crabs from outlying areas can enter and restock these depleted

Investigations during the 1950 season on an unsprayed plot indicated that the crab population of that area increased until the latter part of July after which a fairly stable level was maintained until the middle of September. A low and uniform natural mortality occurred throughout the season. This natural mortality rate gave a basis for comparison with losses on the sprayed areas.

Fiddler Crabs.—The evaluation of DDT mortality to salt-marsh fiddler crabs, Uca pugnax, proved difficult due to their burrowing habits. Losses occurred and in some cases were very great. In general, however, they were proportionately less than those suffered by blue crabs.

Besides general observations, a number of techniques were employed to measure fiddler crab populations and mortality.

The use of square frames of screen or metal, measuring 1 to 2 feet on a side and 18 inches high, proved of most value. These were placed in previously dug holes in banks from which all crabs had been removed. They extended at least 12 inches below the surface, the greatest depth at which fiddler burrows were encountered. After the mud was replaced, a known number of crabs were introduced and a cover of hardware cloth or screening placed on top to prevent escape of fiddlers and depredation by birds. Counts of live and dead crabs dug after spraying indicated mortalities in the different dosage areas.

Another procedure involved the use of a foot-square frame having sides 6 inches high. After this frame was forced a short distance into the ground in selected localities, collections and counts were made of all the small crabs and other invertebrates within its confines. Sampling by this means indicated considerably less mortality to small fiddlers in grassy, more protected areas than in open banks.

Kills in areas treated at dosages of 0.8 pound per acre and above were more immediate than in those treated at lower rates. Many dead and dying crabs were noted in the former areas on the day following spraying, but peak mortalities were noted on the second day. Thereafter, deaths declined and mortality largely ceased 10 days after spraying. The casualties in bare flats were proportionately greater than those occurring on creek and ditch banks where the drainage was better.

In the lower dosage plots, losses were usually first noted on the second day after treatment and were greatest on this or the third day. By the fifth to seventh day after spraying no further mortality from DDT could be observed.

Counts of crabs dug out a week after spraying indicated a total reduction of 42 per cent in the 0.5 area and 24 per cent in the 0.25 area of which 22 and 11 per cent respectively were due to known mortality. At the same time the check area suffered an eight per cent reduction and one per

cent known mortality. Although this method was not used in the higher dosage areas, a similar technique there revealed mortalities up to 50 per cent.

The studies also indicated that losses among small-sized fiddlers less than 0.5 inch wide were greater than those among larger individuals. Mortalities ranged 10-15 per cent higher in the 0.25 area and were complete or nearly complete in the

1.6 plot.

Many affected, sluggish crabs were seen after sprayings. Some of these probably survived but many were eaten by birds as evidenced by the greatly increased numbers of large claws of the males. Comparisons were made of the average numbers of claws found per day along the banks of the enclosed creek and ditch systems in each area. These indicated that mortality from spraying was from 3.5 to 34 times as great as that found either before treatment or after DDT mortality had ceased. The greatest increase (34 times) was observed at the higher dosages.

Some dead crabs were noted also at distances of over 200 yards beyond the boundaries of the sprayed plots. Such observations indicate that as a result of spray drift and the transport of DDT by tidal creeks and ditches, the hazards of treatment are not confined to the limits of the

sprayed plots.

Other Invertebrates.—Soil-surface sampling indicated no observable effects to mussels (Volsella demissa) from DDT applications. A slight seasonal increase in numbers was noted in these studies. Groups of mussels were also located and counted at intervals throughout the summer. No mortality was observed after the August, 1950 spraying. Some deaths occurred in September but the same was also noted in the check area.

Saltmarsh snails (Melampus lineatus) were very common and likewise did not appear to be affected by DDT treatments. Soil-surface sampling showed a general increase in the total numbers and in the total volume of snails in all plots following spraying. In similar manner, the average snail size increased through the

summer in all except the 1.6 plot where increased numbers of small individuals reduced the size average slightly. Although counts in the 1.6 area in 1950 were somewhat below the numbers recorded the previous year, a similar situation prevailed in the check area.

A number of dead and sluggish clam worms (Nereis sp.) were noted on the soft mud bottoms of shallow ponds and drained creeks within a day following spraying. Although such effects were observed in all the study plots, they were most noticeable in the high-dosage areas. Mortality appeared to extend for several days. No quantitative counts were made.

Bait shrimp (Palaemonetes pugio) proved very susceptible to DDT poisoning. Considerable numbers of dead individuals were noted along creek and ditch banks within 12 hours after spraying. These observations were confirmed by effects on shrimp confined in cages as well as by seine hauls made under standard conditions. talities were severe in the 0.5 area and light to heavy in the 0.25 area. However, some live shrimp were always seen after spraying suggesting either that not all were affected or that enough DDT had been washed out to make these waters tolerable. Mortality apparently ceased by the third day following application and more live shrimp were seen thereafter. Two weeks after spraying, they were as plentiful as before treatment. Undoubtedly reinvasion accounted in large part for this repopulation.

Soil-surface sampling revealed clear-cut evidence of the high susceptibility of marsh fleas (Orchestia grillus) to DDT spray. Following the August, 1949 treatment, no live individuals were seen in any of the areas. Second-year observations in the 1.6 plot showed that these amphipods were still absent one full year following spraying. Consequently, in addition to being highly susceptible, it appears that this form is slow to reestablish itself on treated areas. Live marsh fleas were noted after the August, 1950 treatment in a high part of the 0.25 area under a dense mat of dead grass. This indicates that they can survive if, in

addition to protective cover, they are above the normal tide level of DDT-bearing waters. During the two-year period, populations in the check area maintained themselves or increased.

Salt-marsh sowbugs (*Philoscia* sp.) showed DDT susceptibility similar to that of marsh fleas. However, several live individuals noted in soil samples after spraying suggest that they may be more resistant.

Contrary to reports of high resistance by some spiders to DDT, the present investigations indicate these salt-marsh arachnids suffered heavy losses. Within 24 hours of spraying, dead and dying individuals of the commonly distributed wolf spider were noted throughout the treated areas. However, conspicuous numbers of dead spiders were apparent for only two days after spraying.

Results of soil sampling indicated 50 to 90 per cent reductions in the numbers of ground-inhabiting spiders after spraying. The casualties were greatest in areas of heaviest application. Meanwhile checkarea populations remained the same. There was no recovery of spiders in sprayed plots by the end of the first summer but studies in August, 1950, in the 1.6 plot (not treated second year) showed that populations were only a little below the original pre-spray levels.

The effects upon web-building spiders were much the same. Webs inhabited by spiders before spraying were deserted afterwards although similar studies in check areas revealed practically no reduction in number of active webs. Populations sprayed in midsummer did not show any degree of recovery in net-sweeping counts until early the following summer but the effects from treatments in early summer persisted for less than a month.

In contrast to the decline of spiders, there were general increases in red mite populations following spraying. This was revealed by soil samples and by net sweepings. The largest gains were observed in the 1.6 area. Meanwhile check area populations remained nearly stable. Similar large increases of red mites following DDT spraying have been noted elsewhere.

SUMMARY

Experimental studies were conducted on a New Jersey salt marsh during the summers of 1949 and 1950 to determine the hazards to wildlife from mosquito control involving use of DDT. Four plots were established and treated by plane at dosages ranging from 0.2 to 1.6 pounds of DDT per acre. Each plot was sprayed two or three times each year at approximately monthly intervals, except the 1.6 area, treated only in 1949. Studies were conducted before and after each treatment to determine the effects of spraying upon populations of the principal wildlife species and upon invertebrates which serve as food for wildlife.

Birds apparently suffered little direct harm although a one-week old clapper rail died from acute DDT poisoning on the 0.25 area. Local movements of swallows and gulls were noted in response to depletion or increased availability of food but these were only temporary and no mortality was evident. No changes resulting from sprayings were observed in numbers of song birds after the breeding season and they, along with rails, appeared to find sufficient food.

Killifish proved susceptible to DDT spray with heavy kills occurring at the 1.6 and 0.8 pounds per acre applications. Visible effects of spraying continued up to five days after treatment. Only light kills were observed at dosages below 0.5 pound

per acre. However, cage experiments at these same levels indicated considerably higher mortalities to confined fish. Ponds generally sustained greater losses than did creeks and ditches. Repopulation of waterways by fish from outside the areas occurred rapidly except in the case of ponds not affected by daily tide flows.

DDT applications were very toxic to blue crabs. Dosages of 0.8 pound per acre and above resulted in complete or nearly complete kills of the crabs. Such areas remained toxic for two weeks. Lower dosage levels showed population reductions ranging from 20 to 80 per cent over a seven-day period following spraying. Time required for repopulation varied from a few days (0.25 area) to 2 weeks (0.5 area) after mortality ceased. As with fish, crab losses in ponds were higher than those in creeks and ditches.

In common with other crustaceans, fiddler crabs were readily affected by DDT treatments. Losses of 10 to 20 per cent occurred in the 0.25 area and 20 to 40 per cent in the 0.5 area. Somewhat greater mortality resulted at higher dosages. Small fiddlers less than 0.5 inch wide were most susceptible.

Other small invertebrates were variously affected. Amphipods, sowbugs and bait shrimp incurred very heavy losses while insects, spiders and worms were somewhat more resistant. Little or no apparent harm resulted to red mites, snails and mussels.

STORING ADULT MOSQUITOES

Because mold was destroying so many adult specimens of mosquitoes during a recent stay in India, the following methods for storing were developed: r. Flat wooden storage boxes were built by local carpenters. 2. Beeswax, to serve as a substitute for cork, was purchased in the bazaar. 3. The following mixture was melted and poured into the bottom of the box, and allowed to cool, ½ (by volume) beeswax, ½ naphthalene flakes. 4. Any cracks which formed in the cooling process were pressed out. 5. After white paper was glued on the bottom, the box was ready for use.

For the storage of unpinned mosquitoes, metal ointment boxes were used as follows:

1. One quarter inch of flakes was placed on the bottom.

2. The flakes were melted by holding the box over an alcohol lamp and allowed to cool.

3. A small amount of cotton was pressed down over the naphthalene.

4. The heating process was repeated allowing the naphthalene to vaporize through the cotton. Occasionally it was necessary to press down the cotton afterward but only after it was thoroughly cooled.—Charles O. Masters.