Table 2.—Larvicides evaluated in screening tests only.

<table>
<thead>
<tr>
<th>Larvicide</th>
<th>Lowest concentration causing 100% mortality (p.p.m.)</th>
<th>Acute oral toxicity to rats (LD₅₀ mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bay 48792</td>
<td>0.1</td>
<td>2.5</td>
</tr>
<tr>
<td>Bay 48772</td>
<td>0.05</td>
<td>3</td>
</tr>
<tr>
<td>Bay 57553</td>
<td>0.01</td>
<td>5</td>
</tr>
<tr>
<td>Stauffer N-1548</td>
<td>0.05</td>
<td>30</td>
</tr>
<tr>
<td>Stauffer N-1524</td>
<td>0.05</td>
<td>10</td>
</tr>
<tr>
<td>Velisol FCS-303</td>
<td>0.05</td>
<td>Unknown</td>
</tr>
</tbody>
</table>

Bay 48792: (O-ethyl O-[(p-nitrophenyl) - phenyl] ethylphosphonothioate)
Bay 48772: (O-methyl O-[(p-methylsulfonyl)-m- tolyl] methylphosphonothioate)
Bay 52553: (O-isopropyl O-methyl O-p-nitrophenyl phosphonothioate)
Stauffer N-1548: (S-[(p-chlorophenyl)thio] methyl O-methyl methylphosphonothioate)
Stauffer N-1524: (O-ethyl methylphosphonothioate)
O-ester with 2-chloro-1-hydroxybenzonitrile
Velisol FCS-303: (4-(4-bromo-2,5-dichlorophenyl) O-ethyl phenylphosphonothioate)

9098, Bay 77488, CIBA C-9491, Monte
catini L 561, CIBA C-8874, Bay 69047,
and Stauffer N-4988 were about 1.5 to
0.5 times as toxic as DDT. The remaining
compounds were about one-third to one-
seventh as toxic as DDT.
Additional compounds that gave good
kills in screening tests but were not tested
further because of high mammalian toxic-
ity are listed in Table 2. The chemical
composition of each of these compounds
is given in Table 2a.

LARVAL DEVELOPMENT OF Aedes aegypti (L.)
IN USED AUTO TIRES

J. E. KEIRANS

Since the inauguration of the Aedes
aegypti Eradication Program in 1964,
many new studies on the control of this
mosquito have been undertaken, and
much new information on its habits has
been made available. Tinker (1964)
showed that the receptacle most favored
as a larval site by A. aegypti was a used
tire. Haverfield and Hoffman (1966)
found that the shipment of used tires is
probably the most efficient means of dis-
persing this mosquito and that its larvae
are found more often in tires than in any
other type of container. The recognition
of this fact led to a series of experiments to
determine if (1) an auto tire freshly dis-
carded is capable of supporting larval
development of A. aegypti, (2) the length
of the larval development span in tires
of various ages, and (3) the amount of
food available to the larvae in tires of
known age.

MATERIALS AND METHODS. Four sites,
referred to herein as A, B, C, and D, were
selected in and around Savannah, Georgia,
each naturally infested with A. aegypti
and each different in its vegetation and
proximity to organic food sources for the
larvae. All botanical designations are
from Small (1943).

Site A. A small ditch beside a fre-
quently used road, the general vegetation
being Phytolacca americana (pokeweed),
Quercus virginiana (live-oak), Quercus
ilicifolia (scrub-oak), Viola sp. (wild...

1 From the Biology Section, Technical Develop-
ment Laboratories, National Communicable
Disease Center, Health Services and Mental Health
Administration, Public Health Service, U. S. De-
partment of Health, Education, and Welfare,
Savannah, Georgia 31402.
grape), *Pinus australis* (long-leaf pine), *Ambrosia elatior* (ragweed), and *Althaea* sp. (chickweed).

Site B. A 26- x 28-ft. yard enclosed by a 6-5-ft. brick wall behind an abandoned three-story house. Vegetation—*Melia azedarach* (china-berry), *Quercus virginiana* (live-oak), *Eupatorium capillifolium* (dog-fennel), and *Cenchrus tribuloides* (sand-spur).

Site C. A 19- x 41-ft. used-tire yard surrounded by a 6-ft corrugated fence in a standard section of Savannah. Vegetation in this area includes *Albizia julibrissin* (mimosa), *Eupatorium capillifolium* (dog-fennel), *Pharbitis* sp. (morning-glory), *Cenchrus tribuloides* (sand-spur), *Digitaria* sp. (crab-grass), *Amaranthus* sp. (pigweed), *Liquidambar styraciflua* (sweet-gum), and *Platanus occidentalis* (sycamore).

Site D. The northwest corner of a cemetery, 10 yards from a dirt road. Vegetation includes *Celtis* sp. (hackberry), *Liquidambar styraciflua* (sweet-gum), *Miaabiliis jalapa* (four-o’clock), *Pueraria thunbergiana* (kudzu vine), *Ambrosia elatior* (ragweed), *Cirsium* sp. (thistle), and *Holcus lanipes* (Johnson-grass).

Used auto tires were obtained locally and were of various sizes. All had been recently removed from autos and stored dry before experimental use. Before being placed in the field, the inside of each tire was vacuumed to remove any previously accumulated food material.

Initially, 10 tires were placed at each site. They were mounted at a 45° angle by tilting them on stakes, and 2 liters of tap water were added to each tire. The water level was maintained at approximately the 2-liter level by additions, if necessary, but excess amounts of rain water were not removed. To each of two tires at all four sites, 25 newly emerged *Ae. aegypti* larvae were added the same day the tires were placed in the field. Likewise, 25 larvae each were added to another two tires at each site after periods of 1, 2, 3, and 4 weeks. This procedure allowed food materials to accumulate in the tires for specific periods of time before the addition of larvae. The time periods required for larval development in the tires were recorded. Pupae were picked daily and the resultant adults identified to species. The temperature data for the test period were obtained from the U. S. Weather Bureau, Savannah, Georgia.

**RESULTS AND DISCUSSION.** In previous laboratory studies, full rations of a finely ground standard laboratory chow were designated as 0.15 mg. and 0.30 mg. per larva on days 0 and 1, respectively, and 0.60 mg. per larva daily thereafter. Larvae were also reared at 1/2 and 1/4 of these amounts. Using these food regimens, larval development times were determined at constant temperatures of 60°, 70°, 80° and 90° F., and at fluctuating 24-hr cycles of 70° to 75°, 60° to 80°, 70° to 90° and 80° to 100° F. (Keirans and Fay, 1968). An estimate of the accumulated food supply in the tires was then possible by comparing the larval development periods from the laboratory and field tests using comparable temperature conditions.

In tires seeded with larvae immediately (0 weeks), the time required to initial pupation varied from 12 to 18 days and pupation varied from 0 to 140 percent (Table 1). No pupation occurred in one tire, and chemical analysis indicated the presence of soap, which is often used as a lubricant to fit tubeless tires to the wheel rims. The 140 percent pupation was due to additional eggs deposited by naturally occurring *Ae. aegypti*. Larvae added to tires at 1 and 2 weeks varied in initial pupation time from 6 to 9 and 6 to 8 days, respectively. In tires aged 3 weeks, the first pupae appeared on days 5 through 14 (Table 1). In the latter case, since the 14-day figure is so obviously at variance with all the pupation times in the other 3-week-old tires (Table 1), it was not included in the totals. Initial pupation in the 4-week-old tires varied from 5 to 10 days. Tires at Site A, situated in a roadside ditch, were flooded out.

In tires exposed for 0 weeks the average values for initial pupation and temperature at the four sites were 13.3 days and 73° F. In the laboratory the nearest correspond-
Table 1.—Initial pupation (days) for 25 first instar Aedes aegypti placed in pairs of auto tires previously exposed in four field environments for 0, 1, 2, 3 and 4 weeks, respectively. Values for percent pupation shown in parentheses.

<table>
<thead>
<tr>
<th>Field Site</th>
<th>Replicate</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4 flooded</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1</td>
<td>12 (32)</td>
<td>9 (76)</td>
<td>6 (12)</td>
<td>6 (91)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>18 (76)</td>
<td>9 (60)</td>
<td>8 (68)</td>
<td>9 (60)</td>
<td>flooded</td>
</tr>
<tr>
<td>B</td>
<td>1</td>
<td>.. (0)</td>
<td>8 (80)</td>
<td>6 (92)</td>
<td>5 (76)</td>
<td>5 (84)</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>13 (68)</td>
<td>8 (92)</td>
<td>6 (96)</td>
<td>7 (96)</td>
<td>9 (396)</td>
</tr>
<tr>
<td>C</td>
<td>1</td>
<td>13 (144)</td>
<td>9 (68)</td>
<td>6 (152)</td>
<td>.. (0)</td>
<td>7 (176)</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>13 (124)</td>
<td>8 (68)</td>
<td>7 (112)</td>
<td>14 (120)</td>
<td>7 (122)</td>
</tr>
<tr>
<td>D</td>
<td>1</td>
<td>12 (88)</td>
<td>6 (116)</td>
<td>6 (92)</td>
<td>3 (100)</td>
<td>5 (72)</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>12 (80)</td>
<td>6 (100)</td>
<td>7 (104)</td>
<td>9 (92)</td>
<td>10 (104)</td>
</tr>
</tbody>
</table>

a Soap in tire.
b Three Toxorhynchites rivulus larvae (Diptera: Culicidae) and several Copelatus sp. (Coleoptera: Dytiscidae) larvae and one adult S. present. Both predacious on Aedes aegypti larvae.
c Probable malathion contamination confirmed.
d Stolen after 18 larvae had pupated.

...ing pupation time under similar temperatures of 70°F. or 68°F to 80°F. (Table 2) occurred on day 11 at 1/4 food ration. Average pupation times in tires aged for 1 week and in the laboratory tests were approximately equal but the average field temperature was 2°F cooler than the 80°F. in the laboratory, indicating slightly more than 1/4 food ration available to the tire-reared larvae. Similarly, with tires at 2, 3 and 4 weeks, a comparison of temperatures and initial...

Table 2.—Estimated food accumulation in tires exposed to field conditions based on average temperature—time factors to initial pupation and controlled laboratory studies.

<table>
<thead>
<tr>
<th>Field Exposure Period (Weeks)</th>
<th>Average Temp. (°F.)</th>
<th>Average Initial Pupae (Days)</th>
<th>Est. Food Regimen</th>
<th>Laboratory Daily Temp. Cycle (°F.)</th>
<th>Average Initial Pupae (Days)</th>
<th>Actual Food Regimen</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>75</td>
<td>13.3</td>
<td>−½</td>
<td>60°−80°* a</td>
<td>11</td>
<td>¼</td>
</tr>
<tr>
<td>1</td>
<td>78</td>
<td>7.9</td>
<td>+¼</td>
<td>70°−90° a</td>
<td>8</td>
<td>½</td>
</tr>
<tr>
<td>2</td>
<td>80</td>
<td>6.5</td>
<td>+½</td>
<td>70°−90° a</td>
<td>7</td>
<td>½</td>
</tr>
<tr>
<td>3</td>
<td>79</td>
<td>6.8</td>
<td>+½</td>
<td>70°−90° a</td>
<td>6</td>
<td>Full</td>
</tr>
<tr>
<td>4</td>
<td>74</td>
<td>7.2</td>
<td>+Full</td>
<td>60°−80°* b</td>
<td>9</td>
<td>Full</td>
</tr>
</tbody>
</table>

a Time to average initial pupation for a constant median temperature for the range shown fell on the same day.
b Time to average initial pupation for a constant median temperature for the range shown fell on day 10.
pupation times in the field and laboratory with indicated rations in the laboratory shows better than ½ food rations at 2 and 3 weeks and better than full food in tires aged for 4 weeks.

Other mosquitoes were found in the tires. *Culex* sp. were found at all four sites but not in all tires. A total of 4,384 *Culex* larvae were counted but some were not identified to species. At Site B, 1,770 *Culex* larvae were found in one tire that contained a dead sparrow. The next most numerous species found in the tires was *Orthopodomyia signifera*; 40 larvae were collected from two tires at Site A. Thirteen *Aedes triseriatus* were found in one tire at Site D, and five *Toxorhynchites rutilus* larvae were collected, three at Site A and two at Site D.

Organic material available as larval food in addition to debris from the previously mentioned vegetation included Diptera (excluding Culicidae) and Hymenoptera (mostly ants) present in tires at all four sites; Orthoptera, Thysanoptera, Lepidoptera and Aranae at three sites; Collembola, Odonata, Corrodentia, Hemiptera, Neuroptera, and Coleoptera at two sites, and at only one site (D) Isopods were found. All of the above were found dead and were assumed to be an available source of food for the larvae.

The data gathered from this study show that a freshly discarded auto tire containing water is capable of supporting larval development of *Ae. aegypti*. Under natural conditions there would be approximately a week's accumulation of organic matter in the water before any larvae could be found, since a freshly discarded tire would contain no eggs. This fact would make it even easier for larvae to survive, since more than ¼ food ration is available after 1 week. In addition, as can be seen from the percent pupation figures in Table 1, tires of any age containing water are capable of supporting and bringing through to pupation large populations of mosquito larvae.

**Summary.** Evaluations were made of the capability of auto tires of various ages to support the development of *Ae. aegypti* following exposure to field conditions for 0, 1, 2, 3 and 4 weeks. The criteria for food supplies were based on the time to initial pupation and comparisons with laboratory tests using known amounts of food. At 0 weeks, the used auto tires contained less than ¼ the optimum ration of food per larva but were capable of supporting larval development. At 1, 2, 3 and 4 weeks, the food supplies in the tires were +¼, +½, +½ and +full, respectively.

**Bibliography**


