

TABLE I.—Comparative costs—drums vs. bulk.

Base Case—10,000 Gal./Yr. Usage			
Drums—FTL (75 drums or 4145 gallons)		Bulk—10,000 gallons	
F.O.B. Bayonne, N. J.	\$.7000/gal.	F.O.B. Bayonne, N. J.	\$.4000/gal.
Frt. Bayonne—Norfolk	+ .0854	Frt. Bayonne—Norfolk	+ .0880
	<u>.7854</u>		<u>\$.4880/gal.</u>
Less Quantity Discount	— .0800		
Used Drum Value	— .0400		
	<u>\$.6654/gal.</u>		
	\$.6654 per gal. del'd. in drums to Norfolk		
	.4880 per gal. del'd. in bulk to Norfolk		
	<u>\$.1774 per gal.—net savings, bulk vs. drum purchases</u>		

Therefore, based on 10,000 gallon usage per year, a savings of 10,000 x \$.1774 per gallon, or \$1,774.00, is available by bulk purchases over drum purchases.

It was found that a simple above-ground 10,000-gallon storage tank could be installed at a cost of \$2,000. An underground tank of the same capacity would cost \$2,400. Hence, it can be seen that one year's savings would more than pay for the cost of installing storage facilities.

In considering bulk storage of Flit MLO, there are other items which should be considered. No special tanks or special pumps are needed, as the product is noncorrosive.

Test results indicate that Flit MLO may be stored in bulk for long periods of time (6 months to 1½ years) without appreciably affecting its effectiveness. These evaluations also include determinations on the product that was subjected to freeze/thaw cycles with similar results.

Care should be taken to eliminate free water from bulk handling and dispensing equipment. Small amounts of water due to tank "sweating" and high humidity do not cause adverse effects. However, gross contamination in the magnitude of 5 percent or greater may noticeably reduce the larvicidal activity. Free laboratory service is available from the supplier to determine the presence of water in the event that contamination is suspected.

Flit MLO, as stated above, is noncorrosive and has very low acute oral and acute dermal tox-

icity and, therefore, is safe to handle. Its flash point is 270° F as compared to 165° F for Diesel fuel and, therefore, it is not a severe fire hazard.

In Virginia, we have established three bulk storage stations: one at Virginia Beach-Lynnhaven Mosquito Control Commission's yard, one at the Kempsville-Bayside yard, and one in York County. Smaller commissions who may use less than car-load lots get their material from one of these stations, thereby saving considerable over their cost if they purchased in drums.

We believe that Flit MLO is a good larvicide and by developing the bulk purchasing plan, we have reduced the cost significantly.

VIABILITY OF MOSQUITO EGGS PRODUCED BY FEMALE MOSQUITOES DENIED OVIPOSITING SITES¹

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While carrying out experiments with colonized mosquitoes it is frequently necessary to have large numbers of mosquitoes of the same age

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TABLE 2.—Drum handling costs.

\$6.00/drum is industry average for drum handling. Includes: Unloading truck, storage, unloading of drums, product left in drum (2-3 gal.), and ordering.

10,000 gallons in 55-gallon drums=180 drums
 180x\$6=\$1080/year in drum handling costs

and in specific periods of their life cycle. Although colonies produce all stages of the insect's life cycle at one time or another, many species, including *Culex fatigans*, do not demonstrate a synchronous life cycle such as that found in *Aedes taeniorhynchus*. Thus, when large numbers of a given species of mosquito are required at a specific time either more colonies have to be established or the study is carried out with fewer than desirable numbers.

Many factors influence mosquito egg-laying and development. Bates (1947), working with *Haemagogus spegazzinii* mosquitoes in a temperature-controlled environment, noted that more eggs were laid at 25° C than at 30° C and that egg deposition could be induced a day earlier if temperatures were kept at 30° C. Concerning the role of blood in egg production, studies such as those by Gordon (1922) have determined that *Aedes aegypti* required whole blood for egg development. In other studies, various components of blood and species of animals which are the sources of blood were investigated (Woke, 1937a, b) to determine their influence in *Aedes aegypti* egg production. These studies, however, did little to establish a means for synchronizing production of mosquitoes in their various life stages.

In studies designed to separate various species of mosquitoes on the basis of protein constituents the problem of asynchronized availability of eggs, larvae, and pupae became very apparent. For

instance, if the eggs of a given colony of mosquitoes were collected daily over a period of days the required number could be obtained only by holding the first laid eggs under refrigeration. Those eggs held in refrigeration, however, demonstrated a different content of protein when compared to eggs one day old. To overcome this problem it was necessary to synchronize gravid females in their laying of eggs but not at the expense of egg viability.

MATERIALS AND METHODS. One hundred mated nulliparous female mosquitoes were allowed to take a blood meal from mice that were placed in individual cages and provided with a ten percent sucrose sugar solution made available through cotton wicking inserted in Erlenmeyer flasks filled with the solution. Two types of oviposition sites were made available. *Culex fatigans* and *Anopheles maculatus* were provided with open ceramic containers of water and *Aedes aegypti* with a similar ceramic water container which was lined with white filter paper. For each group of mosquitoes four cages were maintained simultaneously as follows: cage 1 had oviposition sites available from the moment of introduction of the mosquitoes; cage 2 had oviposition sites introduced on the fifth day; cage 3 had oviposition sites introduced on the 10th day; and cage 4 had oviposition sites introduced on the 15th day. Eggs deposited in the cages were collected and counted up to and including the

TABLE I.—Egg-laying incidence of *Culex fatigans*.

100 fed females				
Day	Cage #1 H ₂ O—day 1	Cage #2 H ₂ O—day 5	Cage #3 H ₂ O—day 10	Cage #4 H ₂ O—day 15
1	H ₂ O introduced			
2	0 egg rafts			
3	8 live			
4	5 live			
5	1 live			
6	H ₂ O introduced			
6	1 live			
6	20 egg rafts			
7	4 live			
7	11 live			
8	10 live			
8	10 live			
9	3 live			
9	4 live			
10	2 live			
10	4 live			
11	H ₂ O introduced			
11	3 live			
11	6 live			
11	37 egg rafts			
12	4 live			
12	3 live			
13	4 live			
13	3 live			
14	4 live			
14	1 live			
15	3 live			
15	2 live			
15	1 live			
16	H ₂ O introduced			
16	1 live			
16	3 live			
16	1 live			
17	35 egg rafts			
17	1 live			
17	1 live			
18	3 live			
18	0 live			
18	6 live			
18	4 live			
19	2 live			
19	1 live			
19	2 live (1 failed to hatch)			
20	1 live			
20	2 live			
20	0 live			
21	1 live			
21	2 live			
21	1 live			
Total	58	75	57	45

TABLE 2.—Egg-laying incidence of *Aedes aegypti*.

100 fed females				
Day	Cage #1 H ₂ O—day 1	Cage #2 H ₂ O—day 5	Cage #3 H ₂ O—day 10	Cage #4 H ₂ O—day 15
1	H ₂ O introduced			
2	0 eggs			
3	1176 live			
4	925 live			
5	259 live			
6	152 live			
7	202 live			
8	42 live			
9	152 live			
10	67 live			
11	53 live			
12	38 live			
13	147 live			
14	49 live			
15	23 live			
16	13 live			
17	13 live			
18	72 live			
19	157 live			
20	59 live			
21	37 live			
Total	3636	3910	2714	1957

TABLE 3.—Egg-laying incidence by *Anopheles maculatus*.

100 fed females				
Day	Cage #1 H ₂ O—day 1	Cage #2 H ₂ O—day 5	Cage #3 H ₂ O—day 10	Cage #4 H ₂ O—day 15
1	H ₂ O introduced			
2	2452 live			
3	1121 live			
4	116 live			
5	445 live			
6	58 live			
7	8 live			
8	35 live			
9	25 live			
10	0 live			
11	0 live			
12	0 live			
13	0 live			
14	0 live			
15	0 live			
16	0 live			
17	0 live			
18	0 live			
19	0 live			
20	0 live			
21	0 live			
Total	4260	2705	1232	723

21st day after the adult mosquitoes were introduced.

RESULTS AND CONCLUSIONS. Regardless of the species of mosquito or time of ovipositions, all eggs with the exception of a single *C. fatigans* egg raft laid on the 19th day were found to be viable. As indicated in Table 1, most *C. fatigans* egg rafts were laid in the 21-day period when the oviposition sites were provided on the fifth day and the fewest number of egg rafts laid when the oviposition sites were withheld until the 15th day. Almost identical numbers of viable egg rafts were deposited whether the oviposition site was provided on the first or the tenth day. Typically, the greatest abundance of egg rafts were laid the day following introduction of the oviposition site when the site was withheld. A marked change in the total number of rafts produced was seen only in those groups where oviposition sites were withheld more than 10 days.

Table 2 indicates the egg-laying behavior of *Ae. aegypti* mosquitoes when their oviposition sites were made available at various intervals. Similar to the findings with *C. fatigans*, the greatest total number of eggs laid in the 21-day period occurred when the oviposition sites were provided on the fifth day while the fewest eggs were laid when the oviposition sites were withheld until the 15th day. The *Ae. aegypti* were also similar to the *C. fatigans*; however, *Ae. aegypti* deposited significantly fewer eggs during the 21-day period when the oviposition sites were introduced on the tenth day as opposed to introduction on the first day.

Table 3 indicates the egg-laying activity of *An. maculatus*. These mosquitoes deposited their eggs over a much shorter period than did the other two species. As in the case of the *Ae. aegypti*, a significant reduction in total eggs produced was noted when oviposition sites were withheld until the tenth day after the females were introduced into the cages.

SUMMARY. This investigation has shown that (1) the viability of *Aedes aegypti*, *Culex fatigans*, and *Anopheles maculatus* eggs retained in the gravid female is unaffected; (2) the greatest number of viable *Culex fatigans* egg rafts can be obtained at one time by denying the gravid female an oviposition site for 5 days or more; and (3) in the case of *Anopheles maculatus* and *Aedes aegypti* the greatest total number of eggs can be obtained by having oviposition sites constantly available.

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MOSQUITOES AND AGRICULTURE ON OKINAWA¹

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Throughout the history of mankind agriculture has had a significant impact on the population of mosquitoes in any given geographic region. Sometimes, through his agricultural practices man has inadvertently aggravated mosquito problems by faulty irrigation or poor drainage systems; at other times he has greatly reduced the mosquito problem by draining swamp lands and leveling fields to increase his available crop land. Ordinarily such a progression and procession of changes take place over decades if not centuries.

The island of Okinawa is unique in that in the past 25 years it has changed from an almost totally agricultural economy, chiefly dependent on rice, to a semi-industrialized economy. Ready markets for pineapple and sugar which could be grown on this semi-tropical island helped hasten the agricultural transition from rice grown in flooded fields to crops that could be grown on dry surface fields in both hilly and level areas. As the flora on Okinawa changed so did the fauna. The reduction in the total area of standing sun-lit waters of rice paddies required by *Anopheles sinensis* and *Culex tritaeniorhynchus* not only reduced the numbers of these mosquitoes as pests but decidedly aided the Japanese encephalitis and malaria control programs. However, those relatively level fields covered with organic debris and intermittently laced with

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