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Gelled Diet for Screwworm (Diptera: Calliphoridae) Mass Production

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ABSTRACT A rearing system based on a diet gelled with Water-Lock G-400, a synthetic superabsorbent (poly(2-propenamide-co-2-propenoic acid, sodium salt)) (WL), was compared with the standard rearing system (liquid diet suspended in acetate fibers) for the mass production of screwworms, *Cochliomyia hominivorax* (Coquerel). The WL rearing system yielded 2% heavier pupae, 32% higher egg to pupa survival, and required 54% less diet and 88% less labor than the standard rearing system. Other advantages of the WL system include reduced susceptibility to suboptimal environmental conditions and labor practices, and characteristics conducive to centralization and mechanization of rearing procedures.

KEY WORDS Insecta, *Cochliomyia hominivorax*, mass production, gelled diet

THE SCREWORM (*Cochliomyia hominivorax* (Coquerel)) eradication program of the Mexican-American Commission for the Eradication of Screwworm involves one of the largest insect mass production programs in the world. Nearly 400 billion flies have been reared, sterilized, and released since screwworm eradication began on the island of Curaçao in 1954. The current mass production facility, located in Chiapa de Corzo, Chiapas, Mexico, is designed to produce 500 million flies per week. This level of production requires 113 metric tons of dry ingredients to make 820,000 liters of larval diet at a cost of \$170,000 each week (Marroquin 1985). Most of the labor needed for screwworm mass production is dedicated to the larval stage; the quality of care given during this stage is the primary determinant of fly quality. Advancements in larval rearing technology can decrease costs of screwworm mass production and increase quality of the insects produced.

The hydroponic rearing system currently used for screwworm mass production was developed by Gingrich et al. (1971) and modified by Brown & Snow (1979). This rearing system uses a liquid, hydroponic diet composed of dried blood, egg, and milk (or milk substitute), plus formalin and water. Hydroponic diet is suspended in acetate fibers that support the larvae. Larval rearing is divided into two phases. The first phase (initiation) includes egg incubation (8 h) and the first 48 h of larval development. Eggs are placed into small plastic pans (66

by 46 by 9 cm deep) with a carrageenan-based gelled diet (6% dried blood, 3% dried egg, 3% dried milk, 0.1% formalin, 0.25% carrageenan, and 87.65% water) and maintained at 39°C, 75% RH for 56 h. After initiation, larvae are transferred to larger vats (91 by 152 by 4 cm deep) on the rearing floor (35°C, 70% RH). This phase of rearing (finishing) includes the remaining 96 h of larval development. Old diet is vacuumed from the vats every 4 h and replaced with fresh diet. Mature larvae begin to crawl off the rearing vats 48 h after being transferred to the rearing floor and drop into water filled canals. Water-borne larvae are pumped to a central location where they are separated from the water and placed in sawdust to pupate.

The hydroponic rearing system produces high quality insects at a reasonable cost. However, it is labor-intensive and sensitive to the quality of care. Vacuuming and feeding every 4 h require three labor shifts per day. Incomplete removal of waste diet reduces the amount of fresh diet that can be added to the vats and results in contamination of the fresh diet by metabolic toxins, primarily ammonium bicarbonate (Brown & Snow 1978). The aqueous nature of the hydroponic diet permits water soluble waste products to disperse throughout the vats, contaminating unused diet before the larvae begin to feed. This contamination causes larvae to crawl out of the vats before they are mature, reducing production and fly quality. Vacuuming and feeding are done by several work crews dispersed throughout the 4,000 m² larval rearing floor, making supervision and process quality control difficult.

Harris et al. (1984, 1985) tested nine gelling and solidifying agents for use in screwworm larval diets, and six produced larvae of acceptable size (>60 mg [Hightower et al. 1972]). A synthetic superabsorbent (poly(2-propenamide-co-2-propenoic acid, sodium salt)) marketed under the brand name Wa-

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ter-Lock G-400 (Grain Processing Corp., Muscatine, Iowa) (WL) was most promising. The WL diet was comparable both with the meat diet used for research rearing and the hydroponic diet used for mass production. Advantages of this diet included elimination of acetate; diet use, labor, and vulnerability to fluctuations in quality of care (labor sensitivity) were also reduced.

Taylor & Mangan (1987) compared Water-Lock diet with meat diet. Larval size and survival on this diet were equal to, or greater than, those obtained on meat diet and production was more uniform over time than on meat diet. The WL diet reduced the possibility of inadvertently transferring young larvae between rearing containers (interline contamination), was easier to use, less objectionable to workers, and less expensive than meat diet. Water-Lock diet was adopted as the medium for research rearing of screwworms in 1985. Investigations into the use of gelled diets for screwworm mass production began in 1986.

Taylor (1988) compared two gelled diets (WL and carrageenan) with the standard hydroponic diet suspended in cotton for use in the initiation phase of larval rearing. Fifty-six hours after infestation, the number and total weight of larvae per pan and mean larval weight did not differ significantly between the WL and hydroponic diets. Numbers of larvae per pan produced on the carrageenan diet did not differ from numbers produced on the other two diets. However, total and mean larval weight were lower on carrageenan diet. Unfortunately, when initiation pans with WL diet were transferred to finishing vats, waste WL gel clogged the vacuum heads used to remove old hydroponic diet. The accumulation of wastes that resulted from poor vacuuming reduced size and yield of mature larvae (R.G., unpublished data). Carrageenan gel dissolved in warm hydroponic diet and did not affect vacuuming. Diet gelled with carrageenan was adopted for use in the initiation phase of mass rearing in 1986. Taylor (1988) also compared diets gelled with WL and carrageenan for rearing from egg eclosion to crawl-off. Water-Lock diet produced more pupae with heavier mean weights and higher adult emergence than did carrageenan diet.

The purpose of our study was to compare WL gelled diet with hydroponic diet for rearing screwworms to crawl-off under mass production conditions, and to optimize techniques for a WL rearing system.

Materials and Methods

This study involved seven experiments and a pilot production test. In experiment 1, WL gelled diet was compared with hydroponic diet using unmodified production size vats. In experiments 2-7, we examined the effects of blood concentration in the finishing diet, feeding schedule, time in initiation, and larval density with WL rearing. Fi-

nally, a pilot production test was conducted for one year. All tests were done in the screwworm mass production facility located in Chiapa de Corzo, Mexico.

The VF-84 strain of screwworms was used for all tests except the latter portion of the pilot test when the OW-87 strain was used. Larvae were reared in small plastic pans (66 by 46 by 9 cm deep) or large production sized metal vats (91 by 152 by 4 cm deep). Three rooms in the mass production facility were used for rearing. These were the initiation room (39°C, 75% RH), the main rearing floor (35°C, 70% RH) and the pupation room (30°C, 60% RH). The diet consisted of 6% dried blood, 3% dried egg, 3% dried nonfat milk, 0.1% formalin, and 87.9% water for all experiments except where noted otherwise. Water-Lock G-400 gelling agent (1.2%) was added to the diet for the WL diet groups. Diets were prepared by combining dry ingredients and mixing them well before water (35-37°C) and formalin were added with agitation.

Experiment 1. In experiment 1, two or three grams of eggs ($\approx 22,000$ eggs per g; R.G., unpublished data) were placed inside a 50-g ring of meat on the surface of 3 liters of WL diet in a small plastic pan and covered with an inverted Petri dish to inhibit dehydration. Pans were placed in the initiation room (0 h). Petri dishes were removed after 4 h. At 48 h, 2 liters of WL diet were added to each pan. Hydroponic diet groups were transferred to large vats at 56 h. Two initiation pans with 3 g of eggs each were transferred to each hydroponic vat (6 g of eggs per large vat). Vats were lined with 0.4 kg of acetate fibers saturated with 12 liters of hydroponic diet. Six hours after the larvae were transferred, 8 liters of warm water (40°C) were added to each vat to dissolve waste gel from the initiation pans. Water and waste diet were vacuumed off and 8 liters of fresh hydroponic diet were added. Hydroponic vats were vacuumed and diet was added every 4 h for 84 h. During the first 24 h, 8 liters of diet per feeding were placed in the vats. Subsequently 10 liters per feeding were added. A total of 218 liters of diet was used per vat. Vats were vacuumed dry at 148 h to induce the remaining larvae to crawl off.

Two liters of WL diet were added to the WL pans at 48 h. Pans were moved to the rearing floor at 56 h and 15 liters of WL diet were added. At 80 h, WL groups were transferred to large vats (one initiation pan per vat, 2 or 3 g of eggs per vat) and given 35 liters of WL diet (total of 55 liters of diet per vat). Four groups of four vats each were prepared for each treatment. The comparison between WL diet with 3 g of eggs per vat and hydroponic diet with 6 g of eggs per vat was replicated three times in consecutive weeks. Two replicates comparing WL diet with 2 and 3 g of eggs per vat were done during the following 2 wk.

At 80 h, larvae began crawling out of the rearing vats. Trays were situated to collect larvae from

groups of four vats stacked vertically to reduce variation due to vat position. Larvae that crawled off were collected every 4 h until 152 h, when tests were terminated. During the last 2 wk of this experiment, larval samples were also taken at the mass production larvae-water separator. These samples were treated the same as the experimental samples. The total weight of larvae was recorded and a 15-g sample was taken from each collection. Larvae from each sample were counted and placed in a Petri dish (15 cm diameter) containing 100 ml of sawdust. Samples were held in the pupation room. At 28 h after collection, percentage pupation was determined and remaining larvae were discarded. At 6.5 d after collections, pupae were weighed; at 11.5 d, percentage adult emergence was determined. Mean larval weight, percentage pupation, mean pupal weight, number of pupae, pupal yield (number of pupae produced per gram of eggs), percentage adult emergence, and diet use were calculated for each group of four vats. Data were blocked by week for analysis.

Experiments 2-7. Larvae were reared to crawl-off in small pans. For the controls in these experiments, 1 g of eggs was placed inside a 16.5-g ring of meat on 3 liters (three-feeding protocol, experiments 2 and 3) or 1 liter (four-feeding protocol, experiments 4-6) of gelled diet (0 h). Groups that received four feedings were given an additional 2 liters at 32 h. Pans were placed in the initiation room for 56 h and then transferred to the larval rearing floor. At 56 and 80 h, 5 and 12 liters of diet (20 liters total) were added. At 80 h, pans were placed inside larval collection trays with 4 liters of sawdust. Larval pans were discarded at 160 h, when pupae and sawdust were transferred to the pupation room. After 4.5 d in the pupation room, pupae were sieved from the sawdust. The weight and number of pupae in a 50-ml sample and total weight of pupae were recorded for each pan. From these data, mean pupal weight, number of pupae produced per pan, yield, and diet usage were calculated. In experiment 2, the experimental group was fed a diet in which the blood concentration was increased from 6 to 7% for the 56 and 80 h feedings. Four replicates for each treatment were done. In experiment 3, groups receiving three feedings were compared with those receiving four feedings. Each treatment was replicated three times. The schedule with four feedings was used for both control and experimental groups in experiments 4-7. Time in the initiation room was increased to 80 h for the experimental groups in experiment 4. In experiments 5 and 6, experimental pans infested with 1.25 and 1.5 g of screwworm eggs were compared with pans infested at the standard rate of 1 g of eggs per pan. Six replicates were conducted for each treatment in experiments 4-6. In experiment 7, we examined the effect of finishing environment and blood concentration in the finishing diet at two egg densities. Finishing environments were the main rearing floor and the pupation room. Blood con-

centrations in finishing diet were 6 and 7%. Pans were infested with 1 and 1.25 g of screwworm eggs in consecutive weeks. Each treatment was replicated three times.

Pilot Production Test. The final test was a pilot mass production program of the WL diet rearing system. The pilot test was designed to produce 15 million pupae per week. Based upon the results of experiments 1-7, the 6% blood diet was used for initiation and finishing. One hundred and sixty small rearing pans were prepared daily. Each pan was infested with 1.25 g of screwworm eggs, 2 liters of diet were added and the pan was placed in the initiation room. An additional 2 and 6 liters of diet were added to the pans at 32 and 56 h. Pans were transferred to the main rearing floor at 56 h. Ten liters of diet were added at 80 h and the pans were transferred to the pilot test area in the pupation room. Larvae began to crawl off the rearing pans at ≈ 96 h. Larvae fell into trays under the rearing racks where they were collected every 8 h and placed in sawdust to pupate. Total weight of larvae and number of larvae in a 15-g sample were recorded. The number and weight of pupae in 50 ml were recorded 40 h and 6.5 d after collection. These data were compared with quality control data from mass production with hydroponic rearing.

Labor. Labor requirements for the hydroponic and WL rearing systems were estimated by direct observation and timing of representative tasks.

Data Analysis. Data were analyzed with general linear model (PROC GLM) procedure of SAS for Personal Computers, Version 6 (SAS Institute 1987, pp. 549-640).

Results

Experiment 1 compared WL and hydroponic diets using production floor vats for finishing. Results from weeks 1-3, in which WL diet infested with 3 g of eggs was compared with hydroponic diet infested with 6 g of eggs, indicated that hydroponic-acetate diet produced heavier larvae ($F = 51.3$; $df = 1, 20$; $P < 0.0001$) and pupae ($F = 35.78$; $df = 1, 20$; $P < 0.0001$), and higher yield ($F = 66.97$; $df = 1, 20$; $P < 0.0001$) than did WL diet (Table 1). Flies reared on gelled diet had higher pupation ($F = 5.26$; $df = 1, 20$; $P = 0.033$) and adult emergence ($F = 33.69$; $df = 1, 20$; $P < 0.0001$) rates. WL groups used 38% less diet per 10^4 pupae produced ($F = 383.57$; $df = 1, 20$; $P < 0.0001$).

The low weights and survival observed with WL diet along with the negative correlation between pupal weight and number of pupae ($R^2 = 0.82$), indicated that the density on WL diet may have been too high. For weeks 4 and 5, WL diet infested with 3 g of eggs was compared with WL diet infested with 2 g of eggs. Data from larval samples collected from mass production hydroponic rearing at the same time as the experimental collections are presented for comparison. Larvae and pupae

Table 1. Comparison of Water-Lock diet with 2 and 3 g of screwworm eggs per vat, hydroponic experimental (Exp.) and hydroponic mass production (Prod.) vats with 6 g of screwworm eggs; results shown are \bar{x} with SEM in parentheses

Week	Treatment	n	Larval wt, mg	Percent pupation	Pupal wt, mg	10 ⁴ pupae/vat	Yield (10 ⁴ pupae/g eggs)	% Adult emergence	Liters diet/10 ⁴ pupae
1	Hydroponic (Exp.)	4	68.95 (0.31)	98.81 (0.08)	53.94 (0.20)	7.39 (0.13)	1.23 (0.02)	91.47 (0.54)	29.53 (0.53)
	Water-Lock (3 g)	4	67.14 (0.89)	98.92 (0.08)	53.25 (0.69)	2.73 (0.14)	0.91 (0.05)	94.96 (0.46)	20.27 (1.02)
2	Hydroponic (Exp.)	4	68.66 (0.44)	98.91 (0.15)	53.29 (0.27)	7.70 (0.04)	1.28 (0.01)	93.77 (0.65)	28.33 (0.13)
	Water-Lock (3 g)	4	61.93 (0.58)	99.18 (0.21)	49.01 (0.42)	3.42 (0.09)	1.14 (0.03)	95.25 (0.04)	16.14 (0.41)
3	Hydroponic (Exp.)	4	68.62 (0.72)	98.70 (0.28)	53.53 (0.50)	8.59 (0.24)	1.43 (0.04)	93.63 (0.48)	25.45 (0.73)
	Water-Lock (3 g)	4	61.60 (0.67)	99.26 (0.12)	48.53 (0.54)	3.57 (0.07)	1.19 (0.02)	95.80 (0.08)	15.42 (0.28)
Mean	Hydroponic (Exp.)	12	68.74 (0.27)*	98.81 (0.10)*	53.59 (0.20)*	7.89 (0.17)*	1.31 (0.03)*	92.96 (0.43)*	27.77 (0.58)*
	Water-Lock (3 g)	12	63.56 (0.85)*	99.12 (0.09)*	50.26 (0.70)*	3.24 (0.12)*	1.08 (0.04)*	95.34 (0.21)*	17.27 (0.73)*
4	Water-Lock (2 g)	4	68.66 (0.58)	99.17 (0.20)	55.15 (0.49)	2.45 (0.16)	1.23 (0.08)	95.47 (0.46)	22.72 (1.61)
	Water-Lock (3 g)	4	61.34 (0.76)	98.96 (0.19)	49.08 (0.67)	3.17 (0.15)	1.06 (0.05)	96.19 (0.47)	17.47 (0.80)
	Hydroponic (Prod.)	1	61.83 ^a	93.53 ^a	46.24 ^a	4.64 ^b	0.77	85.12 ^a	23.01 ^c
5	Water-Lock (2 g)	4	62.22 (0.41)	99.48 (0.07)	50.12 (0.35)	2.76 (0.06)	1.38 (0.03)	96.66 (0.45)	19.95 (0.43)
	Water-Lock (3 g)	4	55.09 (0.55)	99.25 (0.06)	44.04 (0.41)	4.01 (0.12)	1.34 (0.04)	95.93 (0.22)	13.74 (0.40)
	Hydroponic (Prod.)	1	59.48 ^a	94.66 ^a	45.67 ^a	5.25 ^b	0.87	87.66 ^a	20.38 ^c
Mean	Water-Lock (2 g)	8	65.44 (1.26)*	99.33 (0.11)	52.63 (0.99)*	2.61 (0.10)*	1.30 (0.05)	96.06 (0.37)	21.33 (0.93)*
	Water-Lock (3 g)	8	58.21 (1.26)*	99.11 (0.11)	46.56 (1.02)*	3.59 (0.18)*	1.20 (0.06)	96.06 (0.25)	15.60 (0.82)*
	Hydroponic (Prod.)	2	60.65 (1.18)	94.09 (0.56)	45.95 (0.28)	4.95 (0.30) ^b	0.82 (0.05)	86.39 (1.27)	21.69 (1.31) ^c

Large rearing vats were used for finishing all groups.

*, Means are significantly different ($P < 0.05$). See text for test statistics from analysis of variance.

^a Mean from 18 collections.

^b From production quality control data.

^c 107 liters of diet per vat calculated from production inventory control data for March 1987.

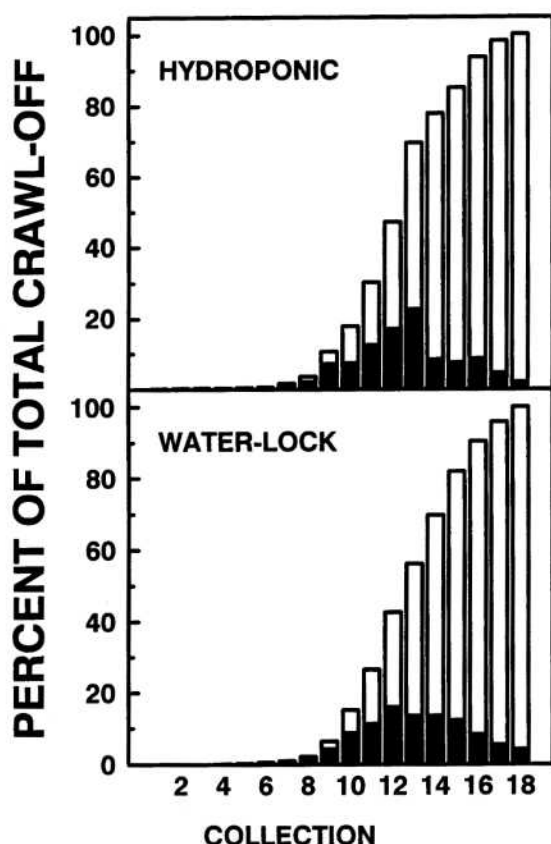


Fig. 1. Percentage of larvae that crawled off and were collected in each 4-h collection period for Hydroponic and Water-Lock (WL) diets in large vats (experiment 1, replicates 1-3). White bars are cumulative.

reared at the reduced density were significantly heavier than those produced at the high density ($F = 163.31$; $df = 1, 13$; $P < 0.0001$ and $F = 147.36$; $df = 1, 13$; $P < 0.0001$) (Table 1). The two egg densities on WL diet did not differ significantly with respect to percent pupation, adult emergence, or yield ($F = 2.52$; $df = 1, 13$; $P = 0.17$; $F = 0.00$; $df = 1, 13$; $P = 0.99$; and $F = 3.86$; $df = 1, 13$; $P = 0.07$, respectively). The pattern of larval development was similar on WL and hydroponic diets (Fig. 1), although the average ages at crawl-off on hydroponic diet (124.4 h) were significantly lower than that on WL diet (126.4 h) ($F = 24.09$; $df = 1, 20$; $P < 0.0001$).

Small pans were used for finishing in experiments 2-7. Preliminary tests indicated that larvae used WL diet more efficiently in smaller, deeper pans. Results of experiments 2-6 are summarized in Table 2. In experiment 2, groups fed standard diet with 6% blood for all three feedings were compared with groups fed standard diet for the first feeding and a diet with 7% blood for the second and third feedings. Results indicated no significant

differences in pupal weight or yield between the two diets ($F = 2.44$; $df = 1, 6$; $P = 0.17$ and $F = 3.80$; $df = 1, 6$; $P = 0.10$). In experiment 3, we compared the standard schedule of three feedings with one with four feedings. The same total amount of diet was used for each treatment. Pupal weight was significantly higher in groups fed four times ($F = 20.08$; $df = 1, 4$; $P = 0.01$). However, fewer pupae were recovered per vat ($F = 13.79$; $df = 1, 4$; $P = 0.02$).

In experiment 4, we compared the standard 56-h period in the initiation room with an 80-h period. Pupal weight did not differ significantly between the two treatments ($F = 2.14$; $df = 1, 10$; $P = 0.17$). However, fewer pupae were produced in the 80 h initiation groups ($F = 24.20$; $df = 1, 10$; $P = 0.0006$). In experiment 5, we compared the standard density of 1 g of eggs per pan with 1.25 g of eggs per pan. The amount of diet was constant for both treatments. Pupae were heavier at the lower density ($F = 12.34$; $df = 1, 10$; $P = 0.006$), but more pupae were produced per pan ($F = 17.10$; $df = 1, 10$; $P = 0.002$) and less diet per 10⁴ pupae produced was required ($F = 14.64$; $df = 1, 10$; $P = 0.003$) at the higher density. Number of pupae produced per gram of eggs did not differ between the two densities ($F = 2.04$; $df = 1, 10$; $P = 0.18$).

In experiment 6, the density in the experimental pans was increased to 1.5 g of eggs. Pupal weight was again higher at the standard density ($F = 30.48$; $df = 1, 10$; $P = 0.0003$), although the number of pupae produced per pan was higher ($F = 17.39$; $df = 1, 10$; $P = 0.002$) and amount of diet per 10⁴ pupae produced was lower ($F = 15.23$; $df = 1, 10$; $P = 0.003$) with the increased density. Number of pupae produced per gram of eggs did not differ between the two densities ($F = 1.62$; $df = 1, 10$; $P = 0.23$).

Diet use averaged ($\bar{x} \pm \text{SEM}$) 12.6 \pm 0.3 liters per 10⁴ pupae produced ($n = 25$) for the controls of experiments 2-6, 16.6 \pm 0.6 ($n = 20$) for WL in the large vats with 3 g of eggs, 21.7 \pm 1.3 ($n = 2$) for hydroponic diet in mass production, and 27.8 \pm 0.58 ($n = 12$) for the experimental hydroponic vats. Mean pupal weights were 48.1 \pm 0.5 mg ($n = 25$) for experiment 2-6 controls, 48.8 \pm 0.7 mg ($n = 20$) for large WL vats infested with 3 g of eggs, 46 \pm 0.3 mg ($n = 2$) for mass production hydroponic vats, and 53.6 \pm 0.2 mg ($n = 12$) for experimental hydroponic vats.

Occasionally, WL diet exhibited excess syneresis during experiments 2-6. The diet became rubbery with free water at the edges of the pans. This condition resulted in high larval mortality. Syneresis was most frequent when the initiation or finishing temperature and humidity were above normal (39°C, 70% RH). Experiment 7 was designed to determine if finishing larvae at a lower temperature and humidity would alleviate this problem. Three parameters—density, percent blood in the finishing diet, and environment during the finishing period—were examined in this experiment. The

Table 2. Comparisons of blood concentration in diet, feeding schedules, time in initiation, and density with Water-Lock gelled diets; results shown are \bar{x} with SEM in parentheses

Experiment	n	Treatment	Pupal wt, mg	No. of pupae $\times 10^4$	Yield (10^4 pupae/g eggs)	Liters diet/ 10^4 pupae
2	4	6% blood	52.32 (0.11)	1.52 (0.01)	1.52 (0.01)	13.1 (0.1)
		7% blood	53.44 (0.71)	1.44 (0.04)	1.44 (0.04)	13.9 (0.4)
3	3	3 feedings	49.18 (0.46)*	1.58 (0.06)*	1.58 (0.06)*	12.7 (0.5)*
		4 feedings	51.57 (0.27)*	1.36 (0.02)*	1.36 (0.02)*	14.7 (0.2)*
4	6	56 h init.	48.41 (0.42)	1.61 (0.06)*	1.61 (0.06)*	12.5 (0.5)*
		80 h init.	49.35 (0.48)	1.30 (0.02)*	1.30 (0.02)*	15.4 (0.2)*
5	6	1 g eggs	46.78 (0.46)*	1.80 (0.06)*	1.80 (0.06)	11.2 (0.4)*
		1.25 g eggs	43.58 (0.79)*	2.12 (0.05)*	1.69 (0.04)	9.5 (0.2)*
6	6	1 g eggs	45.62 (0.82)*	1.48 (0.09)*	1.48 (0.09)	13.8 (0.9)*
		1.5 g eggs	39.22 (0.82)*	2.01 (0.09)*	1.34 (0.06)	10.1 (0.4)*

*. Means are significantly different ($P < 0.05$). See text for test statistics from analysis of variance.

two densities were not tested simultaneously and therefore could not be compared directly.

With a density of 1 g of eggs per pan, finishing environment, percentage blood in the finishing diet, and their interaction significantly affected pupal weight ($F = 8.42$; $df = 1, 8$; $P = 0.02$; $F = 9.51$; $df = 1, 8$; $P = 0.01$; $F = 10.77$; $df = 1, 8$; $P = 0.01$) (Table 3). Pupal weight was highest with the lower finishing temperature and humidity and 6% blood in the finishing diet. Pans with 6% blood in the finishing diet produced significantly more pupae than did pans with 7% blood in the finishing diet ($F = 57.40$; $df = 1, 8$; $P < 0.0001$). Finishing environment and the interaction between environment and blood concentration had no significant effect on the number of pupae produced per pan ($F = 0.08$; $df = 1, 8$; $P = 0.78$; and $F = 0.11$; $df = 1, 8$; $P = 0.75$).

At the increased density of 1.25 g of eggs per pan, heavier pupae were produced at the lower temperature and humidity of the pupation room ($F = 11.54$; $df = 1, 8$; $P = 0.0094$). Blood concentration and the interaction between blood concentration and environment did not have significant effects on pupal weight ($F = 0.00$; $df = 1, 8$; $P = 0.95$; and $F = 0.06$; $df = 1, 8$; $P = 0.81$). Number of pupae per pan did not vary significantly with respect to finishing environment or diet at the high-

er density ($F = 0.44$; $df = 1, 8$; $P = 0.53$; and $F = 2.19$; $df = 1, 8$; $P = 0.18$).

Results of these experiments indicated that the most cost-effective protocol for producing acceptable quality screwworms with WL diet was to infest small pans with 1.25 g of eggs, maintain the pans in the initiation room for 56 h, feed the larvae four times, and finish with 6% blood in the diet at the lower temperatures and humidity of the pupation room. An area of the pupation room was modified for use as a finishing floor for a gelled diet pilot test. Production data for the WL diet pilot test and hydroponic mass production for the year beginning in October 1988 are summarized in Fig. 2. Pilot test production averaged 19.2 million pupae per week with an average pupal weight of 47.1 mg. Yield averaged 1.38×10^4 pupae per g of eggs; 11.7 liters of diet were needed to produce each 10^4 pupae. During the same time period, mass production with the hydroponic rearing system averaged 46.2 mg pupae, 1.06×10^4 pupae per g of eggs and used 25.3 liters of diet per 10^4 pupae.

We estimated the labor required for rearing screwworms with the hydroponic and WL rearing systems by timing representative tasks and by direct observation. Labor requirements for the initiation phase are not presented because the two rearing systems are similar for this stage of the

Table 3. Comparison of Water-Lock gelled diets with 6 and 7% blood finished on the main rearing floor (35°C, 70% RH) or in the pupation room (30°C, 60% RH); values are \bar{x} (SEM) of 3 replicates

Room	Diet	Pupal wt, mg	No. of pupae $\times 10^4$	Yield (10^4 pupae/g eggs)	Liters diet/ 10^4 pupae
1 g eggs/pan					
Floor	6% blood	49.99 (0.99)	1.72 (0.05)	1.72 (0.05)	11.7 (0.3)
Floor	7% blood	50.11 (0.34)	1.23 (0.03)	1.23 (0.03)	16.3 (0.4)
Pupation	6% blood	53.84 (0.40)	1.68 (0.02)	1.68 (0.02)	11.9 (0.2)
Pupation	7% blood	49.87 (0.54)	1.23 (0.11)	1.23 (0.11)	16.5 (1.5)
1.25 g eggs/pan					
Floor	6% blood	45.39 (0.59)	2.07 (0.05)	1.66 (0.04)	9.7 (0.2)
Floor	7% blood	44.92 (1.32)	1.72 (0.26)	1.37 (0.21)	12.2 (1.9)
Pupation	6% blood	50.04 (2.18)	2.03 (0.11)	1.63 (0.09)	9.9 (0.5)
Pupation	7% blood	50.30 (1.32)	1.95 (0.05)	1.56 (0.04)	10.3 (0.3)

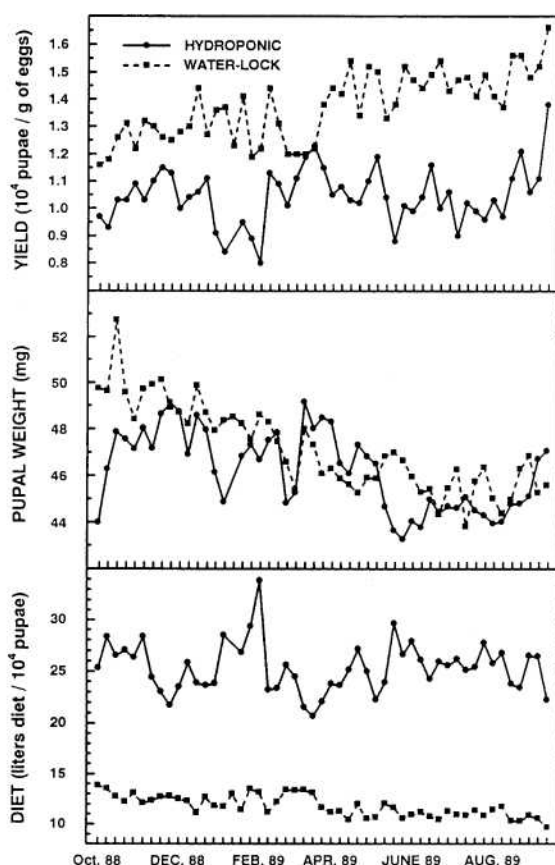


Fig. 2. Pupal yield, mean weight of 5.5-d old pupae, and diet use for hydroponic mass production and WL pilot test. Hydroponic data are from mass production quality control and inventory data.

process. The hydroponic system requires that vats be vacuumed and fresh diet be added 22 times during the finishing phase of larval rearing. Proper removal of old diet requires ≈ 40 s per vat per feeding (5 s per vacuum head application, and eight applications per vat) (Table 4). Refeeding requires an additional 15 s per feeding. Proper waste removal and refeeding requires 55 s per vat per feeding (21 min per vat). Hydroponic vats receiving such care (experiment 1) produced 7.9×10^4 pupae per vat (2.7 min of labor per 10^4 pupae produced). Care given mass production hydroponic vats was suboptimal. At the time of this study (March 1987), vats were vacuumed for an average of 16 s. Each vat received less than 13 min of care. Mass production vats produced an average of 4.9×10^4 pupae during this time period (2.6 min of care per 10^4 pupae produced). The amount of labor per unit of pupae was approximately equal for mass production and experimental hydroponic rearing despite the nearly two-fold difference in labor per vat. Approximately 30 s were needed to feed each pan in the WL pilot test at 56 and 80 h. Pans yielded an average of 1.7×10^4 pupae (35 s of care per 10^4 pupae produced).

Discussion

Results of this study indicate that the WL rearing system can be successfully adapted to screwworm mass production. Compared with concurrent mass production rearing with the hydroponic system, the WL pilot test produced 2% heavier pupae, 32% higher egg to pupa survival, with 54% less diet and 88% less labor per unit of pupae produced. In addition, nearly all of the tasks required for the WL rearing system can be accomplished during one 8-h shift per day, alleviating the need for 24 h per day operation of the mass production facility.

Frequently overlooked in mass production programs is the resiliency, or susceptibility to suboptimal conditions, of the rearing system. Rearing systems must not only perform well under experimental conditions, but must be resilient to the suboptimal environmental conditions or workmanship to which they might be exposed under mass production conditions. The hydroponic system has failed to meet its potential because of its sensitivity to suboptimal workmanship. In experiment 1, hydroponic vats produced 60% more pupae weighing 15% more than concurrent mass production hydroponic vats. This difference can be attributed primarily to labor quality.

Improved labor quality alone cannot overcome the economic advantages of the WL rearing system. The hydroponic system requires more than 2.6 min of care per 10^4 pupae produced. The WL system requires 35 s of labor per 10^4 pupae, and eliminates the need to remove old diet before refeeding, which is the most critical step in the hydroponic system. The WL system permits centralization and mechanization of feeding as well, further reducing labor requirements and vulnerability to labor quality.

The 54% reduction in the amount of diet needed per unit of pupae produced with the WL system offers several potential advantages. The most obvious was reduced expenditures for diet components. Table 5 summarizes the cost of diet components for screwworm larval diets. Although the WL gelling agent is expensive, costing 2.5 times as much per unit of pupae produced as the acetate it replaces, reduced diet use will result in 27% savings in the cost of diet components. At the current level of production (100 million flies per week) a savings of \$9,300 per week or nearly \$500,000 per year can be expected. Lower diet use reduces the amount of organic waste that must be treated and disposed of as well, resulting in lower waste processing costs and reduced environmental impact. Use of waste WL diet as soil conditioners and livestock feed supplements is being investigated.

The WL system requires less space per unit of pupae produced. Because hydroponic vats must be vacuumed and fed every 4 h, adequate space must be maintained above and between vats to allow access. The hydroponic system produces 18.5×10^4 pupae per m^2 of floor space exclusive of aisles (91 by 152 cm vats stacked five high, each pro-

Table 4. Labor for finishing (56 h to crawl-off) with the WL 1.25 g eggs in small pans, 2 and 3 g eggs in large vats, and experimental (Exp.) and mass production (prod.) hydroponic rearing systems

Task	Water-Lock			Hydroponic	
	1.25 g eggs small pan ^a	2 g eggs large vats ^b	3 g eggs large vat ^c	Exp. ^d	Prod. ^e
No. of feedings	2	2	2	22	22
Initial feeding (min:s)	0:30	0:30	0:30	2:00	2:00
Vacuuming (min:s)	0:0	0:0	0:0	0:40	0:16
Subsequent feeding (min:s)	0:30	2:00	2:00	0:15	0:15
Total					
Per vat (min:s)	1:00	2:30	2:30	21:15	12:51
Per 10 ⁴ pupae (min:s)	0:35	0:57	0:42	2:42	2:36

^a WL pilot test, 1.72 × 10⁴ pupae per pan.^b Experiment no. 1, 2.61 × 10⁴ pupae per vat.^c Experiment no. 1, 3.59 × 10⁴ pupae per vat.^d Experiment no. 1, 7.89 × 10⁴ pupae per vat.^e Experiment no. 1, 4.95 × 10⁴ pupae per vat.

ducing 6.3 × 10⁴ pupae). The WL system can produce 62.5 × 10⁴ pupae per m² of floor space (100 by 132 cm mobile racks holding four 66 by 46 cm pans on 12 levels, each pan producing 1.72 × 10⁴ pupae). Because the WL system does not require feeding after 80 h, aisles can be reduced as well. Theoretically, a finishing floor 350 m² could produce 500 million pupae per week. The finishing floor of the mass production facility (designed for this capacity with the hydroponic system) is nearly 4,000 m². Allowing for aisles, rack movement, and proper ventilation, a 75% reduction in floor area should be realized with the WL rearing system. Reduced space translates to reduced construction, maintenance, and energy costs. The WL rearing system eliminates the need for a vacuum system and aquatic larval collecting system. Lower temperatures are allowed on the rearing floor, further reducing construction and maintenance costs for screwworm mass production.

Conversion of the screwworm mass production facility in Chiapa de Corzo, Mexico to the WL rearing system began in October 1989 and was completed in April 1990. The cost for the conver-

sion is estimated to be \$500,000. This includes modifications to the finishing floor, larval collection and diet transport systems, purchase of finishing pans and racks to hold them, diet mixing, dispensing, and waste diet disposal equipment. Savings in diet components alone should pay for the conversion in the first year.

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Table 5. Cost of diet components for gelled and hydroponic diet-rearing systems

Diet	Component, \$ per kg					Total, \$
	Blood, \$0.95	Egg, \$1.54	Milk, \$1.03	Acetate, \$4.73	Water-Lock, \$7.01	
Gelled ^a						
Liter diet	0.057	0.046	0.015	0.000	0.091	0.209
Vat	1.14	0.92	0.31	0.00	1.82	4.20
10 ⁴ pupae	0.66	0.53	0.18	0.00	1.06	2.44
Hydroponic ^b						
Liter diet	0.057	0.046	0.015	0.00	0.00	0.118
Vat	9.01	7.30	2.44	2.47	0.00	21.21
10 ⁴ pupae	1.43	1.16	0.39	0.39	0.00	3.37

Diet and pupal yield data are for the 51 weeks beginning 2 October 1988 and ending 17 September 1989. Costs include purchase price and transportation to the rearing facility in Chiapa de Corzo, Mexico. Price quotes are from October 1989.

^a 20 liters of diet used and 1.7 × 10⁴ pupae per vat.^b 158 liters of diet used and 6.3 × 10⁴ pupae per vat.

ican Commission for the Eradication of Screwworm provided facilities and data for this study.

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