Meridic Diets For Adult Screwworm (Diptera: Calliphoridae)[1, 2]

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ABSTRACT Adults of the screwworm, *Cochliomyia hominivorax* (Coquerel), were fed three carrageenan-gelled meridic diets. In test one, the flies were fed a diet of sucrose and whole dry powdered chicken egg (2:1) and compared with those fed similar formulations containing the sucrose/egg plus nonfat dry powdered milk (2:1:1) or sucrose/egg/milk plus whole bovine blood (2:1:1:2). Body weight, mortality and egg hatch were not significantly ($P = 0.05$) different among the adults. In a second test the mortality and egg hatch were not significantly different between adults fed the sucrose/egg/milk diet gelled with carrageenan or a synthetic copolymer gel. However, significantly greater egg weights were collected from females fed the formulation containing the copolymer gel. In a third test where the copolymer gel was completely or partially replaced in the diet with an inert, water-absorbent material, egg hatch and mortality were not significantly different among the adults. Egg weights were similar for adults feeding on diet containing the full complement of copolymer gel and those fed diet in which the gel was completely replaced with the inert material. These gelled diets are nutritional and economical and have both small scale and mass production application.

KEY WORDS *Cochliomyia hominivorax*, meridic diets, nutrition, adult propagation, colony maintenance, screwworm.

Exogenous protein is not required by the autogenous adult screwworm, *Cochliomyia hominivorax* (Coquerel), for egg development/maturation during the first gonotrophic (ovarian) or egg-laying cycle because larval protein reserves are used for this purpose (Crystal 1966, Mackley and Snow 1982). However, these workers did not mention how quality and quantity of protein in the adult diet affects this particular cycle. Peterson et al. (1987) stated that the exact role of protein in screwworm oocyte maturation is unclear.

In previous tests, a significant increase ($P \geq 0.01$) in total egg weight was observed from female screwworm flies fed a gelled meridic diet used in this study when compared with those females fed only honey (unpublished data). Therefore, this study was undertaken to determine the effects and suitability of use of one or more meridic diets for feeding adult screwworms.

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[2] This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement or recommendation for its use by the USDA.

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Materials and Methods

The study consisted of 3 separate tests. Rearing techniques and procedures for the adults were similar to those used by Taylor (1988a) with some modifications to adapt to our testing procedures. The USDA-ARS Orange Walk-'87 strain of screwworms originally collected in Belize and currently mass produced in the plant facility was used in all tests to insure homogeneity of the flies. This strain had undergone approximately 17 generations (1 year) of colonization.

In each test, five-day old pupae were placed in a 15 × 30 × 12 cm wire cage (W × L × H) until emergence (ca. 1:1 sex ratio) 6-7 days following pupariation. Each cage was provided with 200 ml of diet plus water *ad libitum* and maintained at 26 ± 1°C, 50 ± 10% RH and a photoperiod of 12:12 (L:D). New diet was provided and old diet removed every 72 h from each cage.

Test 1. Three diets were tested five different times using five replicates (cages) per treatment over a period of ca 10 weeks. The control diet contained 27.15% (two parts) sucrose and 13.57% (one part) whole dry powdered chicken egg (S/E) gelled with a 0.52% carrageenan-aqueous solution. The kappa form of carrageenan used was Gelcarin GP-812 (FMC Marine Colloids, Philadelphia, PA). The two treatment formulations were prepared similarly with the exception of one diet containing 13.57% (one part) nonfat dry powdered milk (S/E/M) and the other diet containing the one part milk plus 8.3% spray-dried whole bovine blood (S/E/M/B). (The latter formulation was selected from a previous test in which adults fed similar diets containing final volume blood concentrations ranging from 5 - 50% were compared with adults fed on honey in regards to survival, total egg weight and fertility [unpublished data]).

Fifty ml of pupae (about 500) were placed in each cage (replicate). Parameters measured in Test 1 included body weight, mortality by sex, egg weight and egg hatch (fertility). Body weight determinations were made daily at each testing date for five days following adult emergence from ten males and ten females per cage giving a total of 50 of each sex weighed per treatment. This sampled weight represented 20 percent of a potential population of 2500 flies per testing date (time) for each diet. All weighings were done in a Mettler AE100 analytical balance.

Mortality was expressed as the total number dead by sex per cage during the total time period that the body weights were determined; however, only testing dates two and four were used to determine mortality. Thus mortality represented measurements from a population of a possible 5,000 flies per diet.

Egg weight was expressed as the total quantity (mg) of eggs oviposited within a 30 min time period on lean ground horse meat containing approximately 0.5 ml of spent larval media filtrate. The eggs were collected on day eight following adult emergence from each replicate for testing dates 1, 3, 4 and 5. Therefore a maximum of 1250 females per diet were given the opportunity to oviposit each time or a maximum of 5000 for the total time period.

Each time that the eggs were collected hatch was determined. The fertility represented only those eggs hatched among the total number observed within 24 h following oviposition. The hatched eggs were determined from four mg samples (about 104 eggs/sample) using one sample per cage for each diet.

Test 2. The S/E/M dietary formulation of the first test was gelled using similar aqueous concentrations of the carrageenan or Water Lock G-400 (WL), a

sodium polyacrylate, polyacrylamide polyacrylate copolymer, used in screwworm larval formulations by Harris et al. (1985). The carrageenan gelled diet served as the control diet for the test.

Five replicates (cages) per diet each containing 30 ml of pupae were maintained for this test. Eggs were collected for five consecutive daily ovipositions per cage commencing on the seventh day following emergence and egg hatch was determined from the second oviposition or the eighth day following emergence. Egg collecting techniques and egg hatch measurements were similar to those described in Test 1. The mortality data were collected the same as in Test 1 through day 7 following emergence, thus including total preoviposition time.

Test 3. The WL gelled diet of the second test served as the control diet. Adults fed this diet were compared with those fed three nutritionally similar diets containing a gel replacing or extending material trademarked as grit-o' cobs (GOC) #-60 (The Andersons, Maumee, OH). The GOC material is inert, non-toxic and highly water absorbent. By using final volume GOC concentrations of 3.6, 1.9 and 3.6% the treatment diets were formulated either without WL or only 50 and 80%, respectively, of the WL of the control diet. The same number of replicates and pupae as well as the procedures of data collection for the mortality, egg weight and egg hatch were similar to those used in Test 2 except that the mortality was observed for 11 consecutive days following adult emergence.

Statistical Analyses. The data were analyzed by using analysis of variance (ANOVA) from the SAS ANOVA procedure and mean separations were accomplished with Duncan's Multiple Range Test ($P > 0.05$) (SAS Institute 1988).

Results

Body Weight. In Test 1, no significant differences ($P > 0.05$) were observed in mean body weight for the five day period from the 1250 flies of each sex weighed per treatment (diet) from a possible 6250 flies (Table 1). Nonsignificant daily body weight variations did occur in both sexes on all three diets. For example, a weight increase in general was observed initially but the increase was not sustained and it was not unusual to observe weight loss on the third or fourth day.

Mortality. No significant differences ($P > 0.05$) were observed for mortalities by sex among or between the diets in each of the tests.

Egg Weight. The weight of eggs collected from those females fed the carrageenan gelled sucrose/egg/milk diet in Test 1 was significantly higher when compared with those females fed the other 2 carrageenan gelled diets (Table 1). The pooled egg weights for the 5 ovipositions were higher from females in Test 2 fed this same formulation gelled with WL compared with those fed similar diet gelled with the carrageenan (Table 2). In Test 3 the pooled egg weights for the 5 ovipositions were higher from females fed the control diet compared with those fed diets with WL reduced (Table 3). However, egg weights were not significantly different between females fed the control diet and those fed diet in which the WL was completely replaced with the GOC.

Egg Hatch. No significant differences ($P > 0.05$) were observed for egg hatch among or between the diets in each of the tests.

Table 1. Comparison of screwworm flies fed 3 carrageenan-gelled meridic diets (Test 1).

Parameter	Diet (x + SEM)*			Significance of Diet (ANOVA)		
	Sucrose/egg	Sucrose/egg/milk	Sucrose/egg/milk/blood	F	df	P
Wet wt (mg) - male†	44.1 ± 0.4	43.1 ± 0.3	42.2 ± 0.4	0.68	2,74	0.5657
Wet wt (mg - female†	39.1 ± 0.4	37.3 ± 0.4	36.4 ± 0.5	1.65	2,74	0.1828
Mortality - male‡	21.8 ± 6.0	21.1 ± 5.6	24.2 ± 6.3	0.09	2,29	0.9651
Mortality - female‡	27.4 ± 8.1	22.3 ± 6.5	22.9 ± 6.1	0.11	2,29	0.9565
Eggs (mg)§	100.3 ± 26.7 b	187.4 ± 32.3 a	68.2 ± 15.0 b	11.97	2,59	0.0001
% Egg hatch	72.8 ± 5.0	82.2 ± 3.2	71.6 ± 5.3	1.17	2,59	0.3316

* Means in a row followed by the same letter do not differ significantly ($P = 0.05$; Duncan's Multiple Range Test [SAS Institute 1988]).
† Total of 1250 flies of each sex weighed per treatment from a possible 6250 flies.
‡ Total number dead from a possible 250 flies of each sex per cage (assuming 1:1 sex ratio) within 5 consecutive days following adult emergence.
§ Eggs collected on eighth day following adult emergence.

Table 2. Comparison of screwworm flies fed a meridic diet of sucrose/egg/milk gelled with carrageenan (kappa form) or a synthetic copolymer gel Water Lock G-400 (Test 2).

Parameter	Diet (x + SEM)*		Significance of Diet (ANOVA)		
	0.52% Carrageenan	0.52% Water Lock	F	df	P
Mortality-male†	12.8 ± 1.6	16.2 ± 2.0	1.66	1,4	0.6372
Mortality female†	7.0 ± 0.6	7.2 ± 1.3	4.35	1,4	0.1835
Eggs (mg)‡	810.6 ± 186.1 b	1058.5 ± 55.6 a	11.22	1,4	0.0380
% Egg hatch	86.8 ± 5.1	77.4 ± 5.2	1.06	1,4	0.9544

* Means in a row followed by the same letter do not differ significantly ($P > 0.05$; Duncan's Multiple Range Test [SAS Institute 1988]).
† Total number dead collected within 7 consecutive days following adult emergence from a possible 150 flies of each sex per cage (assuming 1:1 sex ratio).
‡ Total egg weight from 5 consecutive daily ovipositions beginning on the seventh day following adult emergence.

Table 3. Comparison of screwworm flies fed a meridic diet of sucrose/egg/milk using the synthetic copolymer gel Water Lock G-400 (WL) and/or grit-o' cobs #-60 (GOC) (Test 3).

Parameter	Diet (x + SEM)*				Significance of Diet (ANOVA)		
	CONTROL 0.52% WL	3.6% GOC; 0% WL	3.6% GOC; 0.09% WL	1.9% GOC; 0.23% WL	F	df	P
Mortality-male†	35.2 ± 1.5	32.7 ± 2.5	29.1 ± 1.1	32.8 ± 2.5	1.61	3,19	0.2275
Mortality-female†	31.9 ± 3.7	35.5 ± 2.3	32.0 ± 4.3	32.7 ± 1.7	0.28	3,19	0.8384
Eggs (mg)‡	987.0 ± 55.0 a	884.8 ± 28.9 ab	698.9 ± 25.5 c	838.4 ± 29.9 b	10.59	3,19	0.0004
% Egg hatch	86.0 ± 8.0	97.2 ± 1.2	96.6 ± 1.3	90.9 ± 4.0	1.33	3,19	0.2998

* Means in a row followed by the same letter do not differ significantly ($P > 0.05$; Duncan's Multiple Range Test [SAS Institute 1988])

† Total number dead collected within 11 consecutive days following adult emergence from a possible 150 flies of each sex per cage (assuming 1:1 sex ratio).

‡ Total egg weight from 5 consecutive daily ovipositions beginning on the seventh day following adult emergence.

Discussion

Results of Test 1 show that the addition of milk to the egg/sucrose meridic diet will significantly increase fecundity during the first ovarian cycle; however, the addition of whole blood to this formulation did not increase the fecundity. Thus oocyte maturation and number of eggs oviposited are affected by different dietary components prior to the first oviposition in the laboratory. Spradberry and Schweizer (1981) determined that if autogenous females of the Old World screwworm, *Chrysomya bezziana* Villeneuve, ingested dietary protein during the first ovarian cycle they would mature 90% of their oocytes compared with 73% oocyte maturation in females not fed protein for the same time period. These workers state that under field conditions *C. bezziana* females feed on protein during the first ovarian cycle (Spradberry and Schweizer 1979). Thomas and Mangan (1989) observed that the primary reason for wound visitations by female *C. hominivorax* was to feed (62.3% of the time) on the exudates and not to oviposit. Thus the females of this species continually have available to them dietary proteins both of animal origin from wound exudates and those of plant origin from nectar feeding (Mackley and Long 1983).

Data from Test 2 suggest the gel of choice for the adult meridic diet is the WL synthetic copolymer because of its ease of use compared with the carrageenan and observed increased egg weight from those females fed on diet gelled with WL. These gels vary greatly in chemical structure and gelling mechanisms (Taylor 1988b). For example, unlike carrageenan, WL does not require heating in water before mixing with the other dietary ingredients. The results from Test 3 indicate a potential usage of GOC to reduce or replace the WL in the diet and yet maintain a favorable consistency for feeding as well as affording a more economical diet.

The use of a meridic diet has several advantages, e. g., cost, storage, handling and preparation. The dry powdered ingredients used in these meridic diets currently comprise a part of the larval diet. Since this study was conducted, the powdered nonfat dry milk component in the larval diet has been successfully replaced, for economical reasons, with a dry powdered soy-whey material trademarked as Super Fly Starter® (Land o' Lakes, Inc., St. Paul, MN). This material was likewise evaluated in the S/E/M adult formulation in respect to survival, egg mass weight and fertility and was found to be not significantly ($P > 0.05$) different from the diet containing the nonfat milk component.

This diet has been used for colonizing flies collected from Costa Rica in developing a new strain (CR-91) currently being used in mass production for the eradication program. The current cost of honey is about 0.1 cent/g while an equal volume of the S/E/M diet with WL is about one-half this expense.

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