# The need for wheat germ in larval diets of the Mediterranean Fruit Fly, Ceratitis capitata Wied., (Diptera: Trypetidae) of non-nutritive bulking material\*

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### Summary

For normal larval development, the diet of the Mediterranean fruit fly, Ceratitis capitata (Wiedemann), containing pulverized paper as the bulking material required additional nutrients to that of the standard wheat mill feed diet ut lized by the USDA Hawaiian Fruit Flies Laboratory. In addition to increase in dried torula yeast from 3-9%, at least 3% wheat germ was needed. The absence of wheat germ reduced pupal recovery, fecund ty, and egg hatchability of subsequent generations.

# Introduction

Several larval diets of the Mediterranean fruit fly, Ceratitis capitata (Wiedemann), have been formulated and have successfully sustained many generations under mass-production levels. These formulations utilize essentially the same nutrients except that they differ in bulking materials.

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To date, extensively used materials are dehydrated powdered carrot (Mitchell et al., 1965), wheat bran (Nadel, 1965), wheat shorts (Tanaka et al., 1969), and ground sugarcane bagasse (Peleg et al., 1968; Katiyar, 1970). N. Tanaka and associates (unpublished data) have classified bulking materials into 3 classes: nutritive (dehydrated carrot, wheat shorts), less nutritive (wheat bran, coarse mill), and inert (bagasse). A comparison of formulations using the 3 classes of bulking material shows that nutritive materials require 3% yeast level while the less nutritive and inert classes require 6 and 9% yeast, respectively, for normal larval development.

This study was designed to provide training in rearing and to understand the dietary needs of Mediterranean fruit fly larvae cultured on pulverized paper (inert class of bulking material).

# Materials and methods

The standard larval diet of the Hawaiian Fruit Flies Laboratory (Table 1) is based on wheat mill feed, granulated sugar, dried torula yeast (Type 200, St. Regis, Rhinelander, WI), hydrochloric acid, sodium benzoate, nipagin (methyl-p-hydroxybenzoate), and tap water.

In test 1 using the standard formulation, pulverized paper was replaced with wheat shorts, and yeast levels of 3, 6, and 9% were tested to determine the optimum level. Then in test II, the effects of wheat germ in the diet were determined; thus, levels of 0, 1, 3, 6, and 9% (Table 2) wheat germ were tested.

Table -1Formulation of larval diets of milled paper at different yeast levels.

	Weight (g)  Milled paper diets with indicated  % yeast					
Ingredients	Standard	3	6	9		
Sodium benzoate	0.1	0.1	0.1	0.1		
Nipagin (methyl-p-hydro	xy=					
benzoate)	0.1	0.1	0.1	0.1		
Torula yeast	3.6	3.6	7.2	10.8		
Granulated sugar	12.0	12.0	12.0	12.0		
Wheat mill feed	26.0			,		
Milled newspaper		8.1	7.5	6.9		
Hydrochloric acid (ml)	0.2	0.1	0.1	0.1		
Tap water (ml)	58.0	76.0	73.0	70.0		
Total weight (g)	100.0	100.0	100.0	100.0		

	Weight (g)								
	Paper diets with indicated % wheat germ								
Ingredients	Standard	. 0	1	3	6	9			
Sodium benzoate	0.1	0.1	0.1	0.1	0.1	0.1			
Nipagin (methyl-p-hydro	xy=								
benzoate)	0.1	0.1	0.1	0.1	0.1	0.1			
Torula yeast	3.6	10.8	10.8	10.8	10.8	10.8			
Granulated sugar	12.0	12.0	12.0	12.0	12.0	12.0			
Wheat mill feed	26.0								
Wheat germ		0	1.0	3.0	6.0	9.0			
Milled paper		6.9	6.9	6.7	6.5	6.0			
Hydrochloric acid (ml)	0.2	0.1	0.1	0.1	0.1	0.1			
Tap water (ml)	58.0	70.0	69.0	67.2	64.4	61.9			
Total weight (g)	100.0	100.0	100.0	100.0	100.0	100.0			

Mixing procedure: sodium benzoate, nipagin, granulated sugar, and yeast were added to water and mixed thoroughly in 380-ml plastic cups. Then HCl and, finally, the bulking materials were added and mixed thoroughly. pH rate of all diets was ca. 5.5. To each diet, ca. 2500 eggs calibrated volumetrically (Mitchell et al., 1965) were seeded, covered with muslin cloth and kept at temperature of 27°C and 70-80% RH. Each treatment was replicated 5 times. Unlike the paper diets to prevent the larval media of wheat mill feed from drying, 10 ml of water were added to each replication 3-4 days after egg-set. On the 6th day of larval development, the larvae were washed out through the appropriately sized sieve and transferred to the same 380-ml plastic cups containing fine vermiculite for pupation, then sifted 2 days later and the number of pupae recovered were determined by means of wt/no. indices. The percentage pupal recovery was calculated according to egg hatchability. A sample of 100 pupae from each diet was kept in 946-ml cages and adults were provided water and food consisting of a mixture of granulated sugar and enzymatic yeast hydrolysate (4:1). Three egg collections were made for each generation by inserting the 1-fluid-oz plastic receptacles into the cages following 4, 11, and 18 days after adult emergence. Samples of ca. 100-200 eggs from each egging were held on moist filter paper in petri dishes and percent hatch determined.

Pupal size, recovery, egg production and adult eclosion were recorded from 3 generations maintained on each test diet. At the end of the experiment, another 5 replications of  $F_3$  adults reared from the paper diet without wheat germ were fed a mixture consisting of 90% protein hydrolysate and 10% wheat germ oil (Gides, Inc., Long Beach, CA 90813) and compared with adults of the same series that were not offered wheat germ oil.

Evaluation of diets was based on pupal recovery and weight, adult eclosion, and egg production. Analyses of variance were used to note differences among the treatments and the means were ranked by using Duncan's multiple range test.

## Results and discussion

The paper diet required at least 9% yeast for comparable larval growth to that of the standard (3% yeast) diet (Table 3). Diets of 3 and 6% yeast produced smaller pupae and rate of larval development was slower. Larvae began to mature on the 6th day after egg-set, but, at 3% yeast, many were immature.

Table — 3

Comparative effectiveness of paper diets at different yeast levels for rearing larvae of C. capitata (mean±SE).<sup>a</sup>

Paper diets with indicated % yeast	pupal recovery (%) <sup>b</sup>	Pupal wt (mg)°	Adult eclosion (%)°	
Standard	$71.75 \pm 2.39$ c	$7.69 \pm 1.94$	$96.20 \pm 0.86$	
* 3	28.14±2.12 a	$4.41 \pm 0.62$	90.12±1.87	
6	59.64±1.06 b	$5.14 \pm 1.03$	$96.00 \pm 0.89$	
9	75.45±0.83 c	$7.55 \pm 2.23$	$98.00 \pm 0.71$	
V	10.10-0.00 €	1.50 - 2.20	20.00	

<sup>(</sup>a) Each value is a mean of 5 replicates.

<sup>(</sup>b) All paper diets are significantly different at the 5% level (Duncan's multiple range test).

<sup>(</sup>c) Based on random sample of 100 pupae/replication.

There were no significant differences in percent adult eclosion with all diets. When the data were analyzed statistically, the paper diet with 9% yeast gave significantly higher pupal recovery ( $\mathbf{P}=0.05$ ) as compared with other formulations. There were also no significant differences in pupal weight between the standard and 9% yeast level diets. Thus, 9% yeast was used as the base paper diet to test the wheat germ needs, if any, on the larvae.

As shown in Table 4, pupal recovery in the paper diet containing 1% wheat germ decreased with generation. At 9% wheat germ, larval recovery was near equal to the standard diet. However, there were no significant differences (P=0.05) in pupal recovery of diets at 3, 6, and 9% wheat germ levels and the standard diet when the means were compared by analysis of variance. Increase in pupal weights was directly proportional to increase in wheat germ levels. There were no significant differences in adult eclosion in all diets, including the diet with no wheat germ.

Egg production increased with generation with diets containing 3% or more wheat germ (Table 5). When 1 set of  $F_3$  adults reared for 3 generation on diets without wheat germ was fed protein hydrolysate mixed with 10% wheat germ oil, egg production increased markedly, as compared with adults fed without wheat germ oil. Hatchability of eggs also showed a correlation between levels of wheat germ and high hatch.

Studies then showed that diets utilizing non-nutritive bulking materials required 9% yeast for normal larval growth, but to sustain high productive generations of C. capitata the addition of at least 3% wheat germ is required.

The importance of wheat germ was proved better by carrying out a strain at least for 3 generation to determine latent effects of diet.

Table -4Comparative effectiveness of paper diets at different wheat germ levels for rearing of C. capitata (mean $\pm$ SE) (a)

Paper		F <sub>1</sub>			F <sub>2</sub>			F <sub>3</sub>	
diets with indicated % wheat germ	Pupal recovery (%)(b)	Pupal wt(mg)(c)	Adult eclosion (%)(c)	Pupal recovery (%) <sup>(b)</sup>	Pupal wt(mg) <sup>(c)</sup>	Adult eclosion (%)(c)	Pupal recovery (%)(b)	Pupal wt(mg) <sup>(c)</sup>	Adult eclosion (%)(c)
Standard	72±3.34 a	8.16±1.60	97.8±0.37	74±1.00 a	$7.90 \pm 2.69$	$93\pm1.59$	75±1.63 a	$8.70 \pm 2.58$	92.2±1.29
0	55±2.99 b	$5.86 \pm 1.56$	$92.8 \pm 1.46$	44±5.73 b	$6.92 \pm 3.08$	$90 \pm 1.49$	40±1.43 b	$7.86 \pm 1.36$	$89.4 \pm 1.56$
1	64±3.43 ab	$6.41 \pm 1.36$	$94.6 \pm 1.55$	57±6.67 bc	$7.38 \pm 6.67$	$91 \pm 0.67$	50±6.36 b	$7.88 \pm 1.42$	$89.6 \pm 0.24$
3	$68 \pm 2.07$ a	$7.14 \pm 1.07$	$97.4 \pm 0.77$	$67 \pm 4.18$ bc	$7.58 \pm 1.98$	$94 \pm 1.01$	$66 \pm 2.42$ a	$7.90 \pm 1.81$	$96.0 \pm 0.36$
6	72±1.97 a	$7.42 \pm 2.47$	$97.8 \pm 0.45$	69±4.93 a	$7.66 \pm 2.22$	$95 \pm 0.37$	$67 \pm 1.37$ a	$8.18 \pm 2.26$	$93.8 \pm 0.37$
9	$74\pm4.01$ a	$8.16 \pm 1.60$	$99. \pm 0.60$	$75{\pm}1.00$ a	$7.76 \pm 2.01$	$95 {\pm} 0.48$	$78\pm1.63$ a	$8.08 \pm 2.58$	92.6±2.64

- (a) Each value is a mean of 5 replicates.
- (b) Values followed by the same letter are not significantly different at the 5% level (Duncan's multiple range test).
- (c) Based on 100 pupae/replicate.

Table -5 Egg production and percent hatchability of eggs of adult C. capitata reared as larvae on paper diets enriched at different levels of wheat germ.

Paper diets with		$F_1$		F <sub>2</sub>	F <sub>3</sub>	
İndicated % wheat germ	No. of eggs <sup>(a)</sup>	% egg hatchabilit <sup>(b)</sup>	No. of eggs <sup>(a)</sup>	% egg hatchability <sup>(b)</sup>	No. of eggs <sup>(a)</sup>	% egg hatchability(b)
Standard	3541±135	81±2.48	2730±184	86±1.59	3308±140	87 <u>±</u> 1.74
0 .	$1875 \pm 162$	$70 \pm 2.98$	$1840 \pm 133$	$71 \pm 1.13$	$1610 \pm 110$	$65 \pm 4.36$
0°)			•		$2640 \pm 134$	$81 \pm 2.55$
1 .	$2208 \pm 133$	$77 \pm 2.54$	$2480 \pm 144$	$80 \pm 2.62$	$2943 \pm 147$	$84 \pm 2.65$
3	$2625 \pm 120$	$80 \pm 2.06$	$2675 \pm 154$	$82 \pm 3.26$	$2880 \pm 204$	$86 \pm 2.44$
6	$2700 \pm 144$	$82 \pm 2.48$	$2758 \pm 143$	$83 \pm 2.60$	$2966 \pm 160$	$88 \pm 2.64$
9	$2800 \pm 137$	$81 \pm 2.48$	$2983 \pm 214$	$82 \pm 1.86$	$3056 \pm 169$	$87 \pm 1.52$

- (a) Eech value ( $\pm$ SE) is a mean of 3 eggings in a 3-wk period. Calibration: 1 ml = 25,000 eggs.
- (b) Each value ( $\pm$ SE) is a mean of 3 successive egg hatches in a 3 -wk period.
- (c) Adults fed protein hydrolysate containing 10% wheat germ oil.

#### Özet

Akdeniz meyve sineği (Ceratitis capitata Wied.) larva ortamlarında besi değeri olmayan taşıyıcı maddeler kullanıldığında buğday tohumu özü ilâvesinin gereksinimi

C.capitata larva ortamında USDA-Hawa'i Mayve Sinekleri laboratuvarınca, taşıyıcı tampon madde olarak kuilanılmakta'olan ince buğday kepeğ yerine besi değeri olmayan pülverize kâğıtı hamuru kullanıldığında, normal larva gelişimi için ortamdaki torula mayasının %3'ten %9'a çıkarılması gerekmektedir. Maya oranının arttırılmasına paralel olarak kâğıt hamuru ortamına en az %3 oranında buğday tohumu özü (embriyo unu)'nün ilâvesine de gereksinim olduğu saptanmıştır. Bu madden'n yokluğunda birbirini takibeden döllerde yumurta inficar oranı düşmüş ve yumurta verimi ile pupal verim de giderek. azalmıştır.

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