cottonwood (*Populus deltoides* (Bartr.)) nurseries in Mississippi. Report for U.S. For. Serv. Coop. Agree. No. 19-80-374. July-Dec. 1980. 25 pp.

NIELSEN, D. G., F. F. PURRINGTON, AND G. F. SHAMBAUGH. 1979. EAG and field responses of sesiid males to sex pheromones and related compounds. Proc. Symp. Pheromones of the Sesiidae. USDA, SEA, ARR-NE6, p. 11-26.

SHARP, J. L., J. R. MCLAUGHLIN, J. JAMES, T. D. EICHLIN AND J. H. TUMLINSON. 1978. Seasonal occurrence of male Sesiidae in North Central Florida determined with pheromone trapping methods. Florida Entomol. 61: 245-250.

SOLOMON, J. D., AND L. P. ABRAHAMSON. 1972. Hardwood nursery insects. Proc. Southeast. Area For. Tree Nurserymen's Conf., p. 28-33.

SOLOMON, J. D., J. R. COOK, F. L. OLIVERIA, AND T. H. FILER. 1976. Insect and disease canker impact in cottonwood nurseries. Proc. Symp. Eastern Cottonwood and Related Species, p. 301-307.

FEEDING BEHAVIOR AND DIETARY SUBSTRATES FOR REARING LARVAE OF THE CARIBBEAN FRUIT FLY, *ANASTREPHA SUSPENSA*

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Interrelationships between the nutrient and physical composition of larval diets for tephritid fruit flies were first investigated by Mourikis (1965). He discovered that Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), larvae ingest mostly liquid food released from fruit by their mouth hooks and that larvae aggregate at communal feeding sites, presumably to cooperate in liquefying the diet. Many complex diets have since been formulated to mass rear larval fruit flies, such as the Mediterranean fruit fly (Mitchell et al. 1965), *Dacus* spp. (Schroeder et al. 1972), and *Anastrepha* sp. (Spishakoff 1966, Kamasaki et al. 1970). Alternative substrates for these diets have been adopted at different times and places due to the availability of local materials, cost, associated rearing methods and equipment, and assumptions about larval feeding behavior. Consequently, substrates have remained the most variable and undefined of these requirements (Tanaka et al. 1969). The original carrot, sugarcane bagasse, citrus pulp and ground rice hull media have been replaced with diets based on wheat plus an acrylic polymer thickening agent (Tanaka et al. 1969), cottonseed flour with or without a fibrous nylon substrate (Schroeder et al. 1970, 1971), corn cob grits or agar (Burditt et al. 1975), sawdust (Aliniazee & Brown 1977), wheat (Barnes 1979), wheat and soy flour (Schwarz et al. 1985), and corn meal plus agar (Zucoloto 1987). Vargas et al. (1983) compared sugarcane and sugarbeet bagasse, and wheat mill feed as dietary substrates for mass rearing Mediterranean fruit fly larvae. They found no differences in the yields of pupae, although those from sugarbeet bagasse diet were significantly lighter. We conducted this study to determine if larvae of the Caribbean fruit fly, *A. suspensa* (Loew), shred the dietary substrate and ingest particles, and if they can reared on a liquid diet supported by tissue paper, artificial sponge, or powdered cellulose.

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Caribbean fruit fly eggs were derived from a colony established at the Insect Attractants, Behavior and Basic Biology Research Laboratory in 1973 and reared on a corn cob grits diet (Greany et al. 1976). For our experiments, adults were allowed to oviposit for 5 to 6 h periods and the eggs were collected in a 6-oz plastic container by washing them from the substrate with 0.03% sodium benzoate solution. The eggs were rinsed several times with the same solution to remove debris, pipetted onto black filter paper moistened with the solution and incubated for 48 h at 26 ± 1°C. Filter paper supporting 100 embryonated eggs was placed directly in each container of the diet or, after at least 80% of the eggs hatched, 100, 1- to 6-h-old neonate larvae were transferred with a camel's hair brush. All larvae were reared at 26 ± 1°C and 65 ± 15% RH with a 12 h photophase. Pupae were held for emergence in moist vermiculite.

The basic diet was composed of water (400 ml), corn cob grits (Grit-O-Cobs; grade 2040; The Andersons', Maumee, OH 43537; 137 g), sugar (30 g), torula yeast (20 g), raw wheatgerm (15 g), methyl-*p*-hydroxybenzoate (0.7 g), sodium benzoate (0.5 g) and 12 N HCl (2.5 ml). Diet was prepared by first dissolving the sodium benzoate in the water, adding the acid and pouring in the methyl-*p*-hydroxybenzoate dissolved in 5 ml of ethanol. Next, the sugar, finely ground wheatgerm and yeast were added successively. The mixture was then poured into a 4-liter mixer and combined with the corn cob grits. The ingredients were mixed for 5 min at low speed, checked for a pH of 3.5 and dispensed into rearing containers.

Feeding behavior was difficult to observe in the standard corn cob grits diet because the larvae burrowed beneath the surface. Consequently, diets made of white facial tissue dyed with a few drops of red food coloring or commercially available blue and green Kleenex® tissue paper were used. The tissue was shredded in a blender, combined with the other ingredients as before, and dispensed into 15 x 100 mm diam clear plastic petri dishes. Methods used to prepare this diet were similar to those developed for rearing larvae of the apple maggot, *Rhagoletis pomonella* (Walsh), (Neilson 1969) and sorghum shoot fly, *Atherigona soccata* Rondani, (Singh et al. 1983). Each dish received 100, 5- to 6-h-old neonate larvae that were observed from 2 to 5 h after lights-on every day throughout their development. These observations were facilitated by using a color video camera mounted on a stereo microscope.

To test alternative dietary substrates, the corn cob grits were replaced with tissue paper (50 g), cellulose (α-cellulose; Sigma Chemical Co., St. Louis, MO 63178; 150 g), artificial sponge or simply the diet-coated container walls (no dietary substrate). Preliminary evaluations were conducted in 100 ml Erlenmeyer flasks plugged with cotton after being filled with 25 ml of diet and 100 neonate larvae. Final tests were made using 140 x 190 x 20 mm plastic trays containing the filter paper with embryonated eggs. Data were combined from 4 tests, each replicated 3 times per substrate. Parameters included the rate of larval development, number of pupae recovered, individual weights of 120 pupae per substrate (10 per replicate at 2-3 days after recovery), adult emergence and a subjective evaluation of their quality. Means (± SD) were compared by analysis of variance ($p = 0.05$).

The addition of food coloring or use of dyed tissue paper had no apparent effect on larval developmental rates, pupal yields, pupal weights, adult emergence or fecundity. Video recordings indicated that, regardless of source, color was present in the 2nd instar but was most intense during the 3rd instar on day 5. Dyes produced brightly colored larvae, each with an easily visible digestive tract but no color was evident in the tissue or fat body. Prepupae, pupae and adults were colorless, probably because the dyes were excreted (Sharp & Ashley 1984).

Caribbean fruit fly larvae ingested both liquids and solid particles. This was accomplished by means of a feeding mechanism similar to that of the Mediterranean fruit fly (Mourikas 1965). We observed that neonate larva obtain food from the surface of the

diet, perhaps because they lack the anterior stigmatic plates and spiracles developed in the second instar that would allow them to remain partially submersed. By the third instar, fully developed larvae burrowed throughout the diet. All larval stages fed by means of highly sclerotized, paired mouth hooks that were cast back and forth to shred the medium. Ingestible liquids and particles were directed into the suctioning pharynx, a process presumably assisted by the secretion of saliva. Larvae fed on the diet without a supporting substrate but they apparently could not pupate, since most of them drowned during the prepupal stage.

Larvae reared on the paper, sponge, alphacel and corn cob grits diets had similar developmental rates, pupal yields, pupal weights and adult yields. The first pupae appeared on days 7 and 8, and most of the larvae pupated by day 10. Mean (±SD) pupal yields and weights were not significantly different on the respective diets. Yields averaged 34±18, 21±17, 41±30 and 41±30%, and the weights were 16.5±0.6, 15.0±0.9, 13.5±2.8 and 14.9±0.7 mg. Adult yields from these pupae were 89±9, 76±17, 74±17 and 85±9%; significantly higher for paper and corn cob grits.

The frequency distribution in weight categories ranging from 12-20 mg of 120 pupae from each diet indicated that all substrates produced a reasonable proportion of 14-16 mg pupae (Fig. 1). However, the alphacel diet resulted in significantly smaller and the paper larger pupae. The respective diets produced 1, 14, 27 and 7 pupae less than 12 mg. Using 14 mg as a threshold, 96, 72, 63 and 75% of the pupae from the respective diets were of acceptable weight. This is realistic, since Leppla et al. (1976) and Greany et al. (1976) repectively produced pupae with statistically equivalent mean weights of 12.7±1.7 mg and 13.2±0.2 mg on a sugarcane bagasse diet.

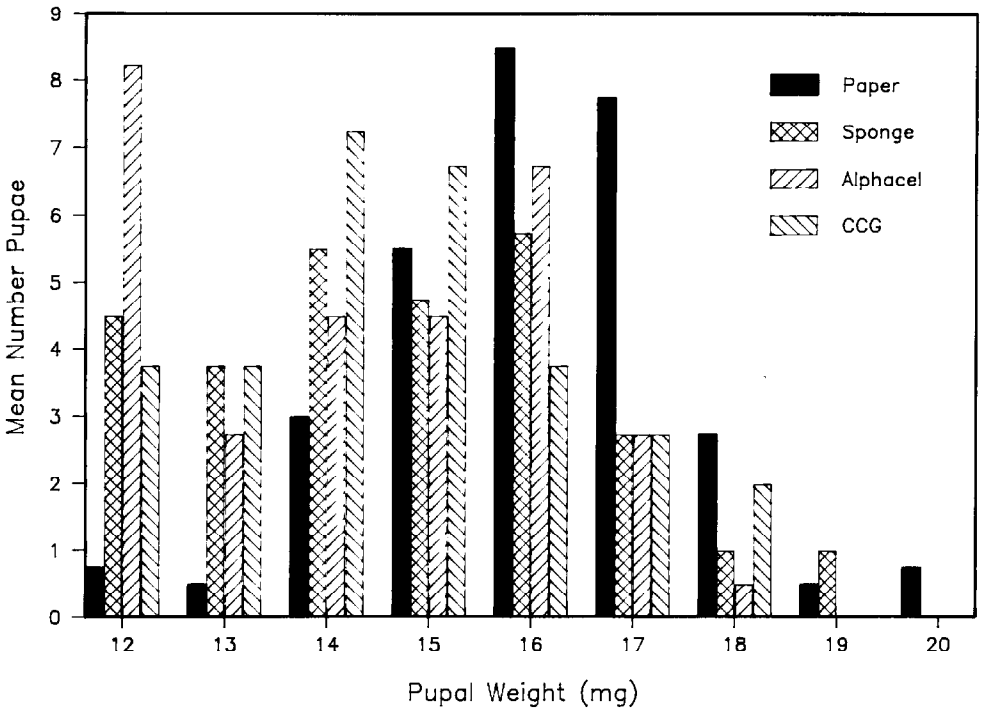

Fig. 1. Frequency distribution of the weights of Caribbean fruit fly pupae from larvae reared using tissue paper, sponge, α-cellulose or corn cob grits as dietary substrates. Of 120 pupae randomly sampled from each diet, 119, 106, 93 and 113, respectively, weighed 12-20 mg.

We conclude that Caribbean fruit fly larvae shred their dietary medium and perhaps require both liquid and particulate food. Ingestible substrates seem to produce larger pupae and increase survival (Zucoloto 1987), so nutritionally inert particles may be important for the regulation of feeding, digestion and absorbtion (Bignell 1978). Remarkably, Carroll (1986) found that Caribbean fruit fly larvae could survive and develop into reproductive adults on a diet of conspecific eggs, neonate larvae and probably associated microorganisms. Thus, virtually any substrate may be used that supports the larvae and diet, remains free of contamination and is practical. It is also desirable for the substrate to be inexpensive, readily available, easy to store and use, consistent, defined in composition and disposable or recycleable. Selection of a substrate for rearing fruit fly larvae, as with any other dietary ingredient, depends more on the quality of fly produced than on economics or expediency.

Acknowledgements

C. O. Calkins and R. R. Rutter provided the eggs, A. L. McAshan handled the pupae, D. A. Carlson and J. Hector produced the figure, and P. D. Greany assisted with the video photography. P. D. Greany and T. R. Ashley contributed particularly helpful manuscript reviews. Mention of a commercial or proprietary product does not constitute an endorsement of that product by the U.S. Department of Agriculture.

References Cited

Aliniazee, M. T., and R. D. Brown. 1977. Laboratory rearing of the western cherry fruit fly, *Rhagoletis indifferens* (Diptera: Tephritidae): oviposition and larval diets. Canadian Ent. 109: 1227-1234.

Barnes, B. N. 1979. Alternative larval media for mass-rearing the Natal and Mediterranean fruit flies. J. Ent. Soc. South Africa 42: 55-56.

Bignell, D. E. 1978. Effects of cellulose in the diets of cockroaches. Ent. exp. & appl. 24: 54-57.

Burditt, A. K., Jr., F. Lopez-D., L. F. Steiner, D. L. Von Windeguth, R. Baranowski, and M. Anwar. 1975. Application of sterilization techniques to *Anastrepha suspensa* Loew in Florida, United States of America, pp. 93-101. *In* Proc. Sterility Principle for Insect Control 1974. IAEA, SM-186/42, Vienna. 622 pp.

Carroll, J. F. 1986. Caribbean fruit flies, *Anastrepha suspensa* (Loew) (Diptera: Tephritidae), reared from eggs to adults on cannibalistic diet. Proc. Ent. Soc. Washington 88: 253-256.

Greany, P. D., T. R. Ashley, R. M. Baranowski, and D. L. Chambers. 1976. Rearing and life history studies on *Biosteres* (*Opius*) *longicaudatus* (Hym: Braconidae). Entomophaga 21: 207-215.

Kamasaki, H., R. Sutton, F. Lopez-D., and A. Selhime . 1970. Laboratory culture of the Caribbean fruit fly, *Anastrepha suspensa*, in Florida. Ann. Ent. Soc. Amer. 63: 639-642.

Leppla, N. C., M. D. Huettel, D. L. Chambers, and W. K. Turner. 1976. Comparative life history and respiratory activity of "wild" and colonized Caribbean fruit flies (Dipt.: Tephritidae). Entomophaga 21: 353-357.

Mitchell, S., N. Tanaka, and L. F. Steiner. 1965. Methods of mass culturing melon flies and Oriental and Mediterranean fruit flies. USDA Tech. Bull. ARS-33-104.

Mourikis, P. A. 1965. Data concerning the development of the immature stages of the Mediterranean fruit fly *Ceratitis capitata* (Wiedemann) (Diptera: Trypetidae) on different host-fruits and on artificial media under laboratory conditions. Annales de l' Institut Phytopathologique Benaki 7: 59-105.

NEILSON, W. T. A. 1969. Rearing larvae of the apple maggot on an artificial medium. J. Econ Ent. 62: 1028-1031.
SCHROEDER, W. J., R. MIYABARA, AND D. L. CHAMBERS. 1970. A reusable diet support for rearing the melon fly on fluid larval medium. J. Econ. Ent. 63: 2002.
SCHROEDER, W. J., R. Y. MIYABARA, AND D. L. CHAMBERS. 1972. Protein products for rearing three species of larval Tephritidae. J. Econ. Ent. 65: 969-972.
SCHROEDER, W. J., R. Y. MIYABARA, N. TANAKA, AND D. L. CHAMBERS. 1971. A fluid larval medium for rearing the melon fly. J. Econ. Ent. 64: 1221-1223.
SCHWARZ, A. J., A. ZAMBADA, D. H. S. OROZCO, J. L. ZAVALA, AND C. O. CALKINS. 1985. Mass production of the Mediterranean fruit fly at Metapa, Mexico. Florida Ent. 68: 467-477.
SHARP, J. L., AND T. R. ASHLEY. 1984. Chemicals tested as internal dye markers for the Caribbean fruit fly, *Anastrepha suspensa* (Loew) (Diptera: Tephritidae). Florida Ent. 67: 575-577.
SINGH, P., G. C. UNNITHAN, AND A. G. L. DELOBEL. 1983. An artificial diet for sorghum shootfly larvae. Ent. exp. & appl. 33: 122-124.
SPISHAKOFF, L. M. 1966. Laboratory rearing of *Anastrepha serpentina*. J. Econ. Ent. 59: 1010-1011.
TANAKA, N., L. F. STEINER, K. OHINATA, AND R. OKAMOTO. 1969. Low-cost rearing medium for mass production of Oriental and Mediterranean fruit flies. J. Econ. Ent. 62: 967-968.
VARGAS, R. I., H. CHANG, AND D. L. WILLIAMSON. 1983. Evaluation of a sugarcane bagasse larval diet for mass production of the Mediterranean fruit fly (Diptera: Tephritidae) in Hawaii. J. Econ. Ent. 76: 1360-1362.
ZUCOLOTO, F. S. 1987. Feeding habits of *Ceratitis capitata* (Diptera: Tephritidae): Can larvae recognize a nutritionally effective diet? J. Insect Physiol. 33: 349-353.

SEASONAL OCCURRENCE OF *GNOPHOTHRIPS FUSCUS* (THYSANOPTERA: PHLAEOTHRIPIDAE) ON SLASH PINE IN FLORIDA

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Adults of the slash pine flower thrips, *Gnophothrips fuscus* (Morgan), feed from Dec-Feb on female slash pine, *Pinus elliottii* Engelm. var. *elliottii*, flowers, so reducing potential seed yields (Ebel 1961, DeBarr 1969, Hedlin et al. 1980). Adults also have been observed from Apr-Sep on current-year's shoots of slash pines of all ages and within the needle fascicles of seedlings (Hedlin et al. 1980). The immature life stages of *G. fuscus* have been described by Ranasinghe et al. (1985). We report on the occurrence of all life stages on young slash pine seedlings and of flying adults within the crowns of older (cone-bearing) slash pine trees in NE Florida.

One hundred young (2-5-yr old) slash pine seedlings growing in the understory of open-grown, mixed slash and longleaf, *P. palustris* Mill., pine trees in a seed production area near Olustee, FL were sampled from Apr 1979-May 1980. At 1-2-week intervals from Apr-Sep and monthly otherwise, an entire current year's (or most recently formed) shoot was collected from each of 15 randomly-selected seedlings. The numbers of thrips life stages present per 15-shoot sample, determined by microscopic examination, were plotted against collection dates.