

ARTIFICIAL DIET AND REARING METHODS FOR THE *MELALEUCA QUINQUENERVIA* (MYRTALES: MYRTACEAE) BIOLOGICAL CONTROL AGENT *OXYOPS VITIOSA* (COLEOPTERA: CURCULIONIDAE)

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The mass production of insects for biological control of pests has become a common technique for permanent establishment, periodic colonization, or inundative releases of agents (Etzel and Legner 1999). For biological control of weeds the production of agents for mass release has been modest possibly due to a lack of knowledge of suitable mass-rearing techniques. Numerous artificial diets have been described for phytophagous insects, especially for pest species of the Noctuidae and Curculionidae (Singh 1977). Although artificial diets have been described for a few species of weed biological control agents (Baer and Quimby 1981; Smith and Wilson 1995; Blossey et al., 2000), none are currently available commercially. The purpose of this study was to develop an artificial diet and rearing conditions for the Australian weevil *Oxyops vitiosa* Pascoe (Coleoptera: Curculionidae) introduced in south Florida (Center et al., 2000) for the biological control of the invasive species *Melaleuca quinquenervia* (Cav.) S. T. Blake (Myrtaceae).

Several artificial diet recipes were tested initially (e.g., Vanderzant 1973; Chang and Jensen 1972; Baer and Quimby 1981; Toba et al. 1969), however, a modified Harley and Willson diet (Harley and Willson 1968) resulted in the best larval survival and performance. These recipes frequently include material from the insect's host plant. For the melaleuca herbivore *O. vitiosa*, each diet was formulated with *M. quinquenervia* tip leaves in various concentrations and modifications were based upon insect performance in previous trials. Tip leaves were blended into a slurry with the described amount of water (Table 1). The diets also were modified by omitting antibiotics and formaldehyde. All diets were dispensed into 1 oz cups and each was covered with a plastic cap. Larvae reared on *M. quinquenervia* plants were obtained from our colonies and tested individually on experimental diets. All test insects were reared at 27°C, 90% RH, and under a 14:10 h photoperiod. During the initial diet screening 3rd instars were tested and if the results indicated that larval feeding, growth and development occurred then 1st instars were tested in subsequent trials.

Third instar larvae were reared successfully (50% survival) through to the pupal stage on diet 1 (Table 1). The adults that resulted from the lar-

vae fed this diet had greater weights than those reared on plant material. Males weighed 39.1 ± 0.3 mg and females 48.0 ± 0.5 mg compared to plant-fed males (36.0 ± 0.5 mg) and females (41.8 ± 0.4 mg). However, when neonate larvae were reared on diet 1, none survived to the pupal stage. Several modifications of diet 1 resulted in 10 additional formulations. The concentration of sorbic acid and methylparaben were decreased while maintaining a culture apparently free of contamination. Additionally, increased amounts of *M. quinquenervia* leaf material were added in diets 10 and 11. Although the neonate larvae fed well on several diets, overall larval survival, prepupal weight, and development rate to the pupal stage were greatest when fed diet 11.

Pupation substrates were evaluated which included mixtures of: 1) all purpose sand (400 g; Bonsal Co., Charlotte, NC, USA), peat moss (56.7 g; Scotts Co., Marysville, OH, USA), and deionized water (100 ml, N = 39); 2) crushed floral foam (42.5 g; Smithers-Oasis Co. Kent, OH, USA), sand (400 g) and deionized water (100 ml, N = 49); and 3) crushed floral foam (42.5 g), sand (400 g), and CuCl_2 (1% in 100 ml deionized water, N = 114). Each substrate was poured individually into 1 oz cups. Prepupae were collected on live plants from field sites and placed individually on each test substrate. All prepupae were reared as described above and the number of emerging adults was counted.

The results indicated that percent survival of the *O. vitiosa* prepupae to the adult stage was relatively high with little difference among the substrates. Adult emergence on the sand/peat moss mixture was 92.3%, 91.8% on the crushed floral block/sand, and 87.7% on crushed floral block/sand with CuCl_2 (1%). These levels of adult emergence are considerably higher than those obtained previously from sand alone (<10%; Wheeler unpublished data) and by others (52%; Purcell and Balciunas 1994) possibly because sand alone was a poor pupation substrate.

SUMMARY

The laboratory colonies of the melaleuca herbivore *O. vitiosa* that were established on plants and those produced on artificial diets yielded a

TABLE 1. ARTIFICIAL DIET INGREDIENTS AND THE RESULTS OF FEEDING NEONATE *O. VITIOSA* LARVAE.

Ingredient	Diet										
	1	2	3	4	5	6	7	8	9	10	11
AlphaCel (g)	11.6	11.6	11.6	11.6	11.6	11.6	11.6	11.6	11.6	11.6	11.6
Sucrose (g)	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6
Glucose (g)	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6	2.6
Corn Starch (g)	3.38	3.38	3.38	3.38	3.38	2.37	2.37	2.37	2.37	2.37	2.37
Wesson Salts (g)	0.775	0.775	0.775	0.775	0.775	0.775	0.775	0.775	0.775	0.775	0.775
Cholesterol (g)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Linseed Oil (ml)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Lecithin (g)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Casein (g)		6.66	6.66	6.66	6.66	6.66	6.66	8.66	8.66	8.66	8.66
Vanderzant Vitamins (g)	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Ascorbic Acid (g)	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Sorbic Acid (g)	0.35	0.32	0.26	0.2	0.16	0.12	0.16	0.08	0.16	0.16	0.16
Methyl Paraben (g)	0	0.32	0.26	0.2	0.16	0.12	0.16	0.16	0.25	0.16	0.16
Agar (g)		5.285	5.285	5.285	5.285	5.285	5.285	5.285	5.285	5.285	5.285
Water (ml)		200	200	200	200	200	200	200	200	200	200
Melaleuca leaves (g)	70	70	70	70	70	70	70	70	70	100	130
N		8	12	31	20	20	93	48	45	44	46
Percent survival to:											
pupa		0	0	0	0	33.3	41.9	35.4	35.6	54.6	60.9
Weight of:											
prepupa						44.1a	44.0 a	45.1 a	49.7 a	44.4 a	50.8 a
se						1.4	1.4	1.4	2.1	1.3	2.1
Development time to: ¹											
pupa						27.0 ab	29.4 a	25.5 b	23.7 b	29.2 a	23.9 b
se						0.8	0.9	1.2	1.2	0.7	0.7

¹Values in a row followed by the same letter are not significantly different according to a Ryan's Q mean comparison test (P < 0.05).

surplus of insects that were released at *M. quinquenervia* infested sites throughout south Florida. In the laboratory, we were able to rear *O. vittosa* from egg hatch through to pupation on artificial diets modified with host plant tissues. The weight gain and development rate of the artificial diet-reared insects was slightly lower but comparable to that of insects fed *M. quinquenervia* leaves (Wheeler unpublished data). For example, about 61% (Table 1) of the diet 11-fed larvae survived to the prepupal stage while 30-90% (N = 20) survived when fed *M. quinquenervia* leaves (Wheeler unpublished data). Mean weight of diet 11-reared prepupae was 50.8 mg (Table 1) whereas leaf-reared prepupae weighed between 59.6 to 66.3 mg. Moreover, the larvae completed development to the pupal stage after 23 to 24 d when fed diet 11 compared with 21 to 23 d when fed leaves. Finally, survival of pupae to adult exceeded 90% when reared in a combination of sand/peat moss and water. Florida Agricultural Experiment Station Journal Series No. R-07918

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