EFFECTIVE MASS PRODUCTION OF EGGS OF THE ANGOUMOIS GRAIN MOTH, SITOTROGA CEREALELLA (OLIVIER).[1/2/]

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ABSTRACT

A production system for eggs of the Angoumois grain moth, Sitotroga cerealella (Olivier), was based on a single insect generation use of wheat. During the same 12-wk period over 3 yrs of egg production, ca. 600,000 eggs were produced/kg of wheat used. A mean weight return of eggs from adults produced was 18.1%. The infestation rate was 39 kg wheat/day. A cost analysis of the system showed the production cost of eggs to be ca. $.00655/1000.

INTRODUCTION

Eggs of the Angoumois grain moth, Sitotroga cerealella (Oliver), serve as an adequate and inexpensive host material for some egg parasites and predators (Morrison and King, 1977). A single insect generation use of wheat for production of eggs of S. cerealella (Morrison and Hoffman 1976 and King et al. 1978) has been predictable, reliable and efficient over 11 yrs of uninterrupted production.

Arthropod contamination encountered in multi-generation systems (Flanders 1930, Spencer et al. 1935) was eliminated in this system and dependable, long-range predictions of production can be made. Further expansion and improvements of the basic rearing system (Morrison and Hoffman 1976) into a functional mass production system are reported.

MATERIALS AND METHODS

Hard red winter wheat with a protein content of 13-15% was used as the rearing medium. Before use, a simple bioassay of the wheat was made to detect pesticide contamination. First, ca. 100 g of the wheat was placed into each of four 250 ml wide-mouthed Mason® jars. Then ca. 1 g of freshly collected adult S. cerealella (ca. 250 moths) was placed into each jar and the jars lidded. The jars were held for 7 days and observed daily for the following criteria: adult mortality (no measurable mortality for 2-3 days), oviposition (large numbers of clustered eggs), egg hatch (normal color progression from cream to red with actively emerging larvae), and larval penetration (larvae actively penetrating grain). After the 7 days, wheat kernels were examined for living larvae. No controls were used because previous experience with pesticide contaminated grain had shown it rapidly

1/ Lepidoptera:Gelechiidae

2/ Mention of proprietary product does not constitute endorsement by the United States Department of Agriculture. This research was done in cooperation with the Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843.

killed the adults and larvae and reduced oviposition. After this determination, the wheat was used for production. All wheat and production equipment were heat treated at ca. 66°C for 24 h before being taken into the rearing areas. This eliminated arthropod contamination and prevented germination of the grain. When cool, 9.1 kg of the wheat was put into each crib.

The rearing cribs measured 2X61X122 cm and were constructed of steel hardware cloth (10 mesh/2.54 cm) bent in a "U" shape around a formed steel tube of 2 cm diam. electrical conduit (Fig. 1A). The hardware cloth was then riveted to the tube at ca. 5 cm intervals. Bolts were placed through both layers of hardware cloth at stress points to keep the wheat in the cribs at a uniform thickness. Once filled, the cribs were thoroughly sprayed with water to restore the wheat to a favorable moisture content (ca. 15%) that was lost during the heating process (Wilkinson and Morrison 1972). The cribs were allowed to drain (ca. 5 min.) and then were placed into a horizontally positioned production module designed to hold 10 cribs (Fig. 1B). The production module (63.5X122X183 cm) was constructed of 1.9 cm exterior grade plywood. The top edge of a sloped metal trough (61X119.4 cm with a 40 cm drop along the length) placed inside the module was bolted at 20 cm intervals to the sides of the module 45 cm from the bottom. A 7.5 cm diam. collection tube 5 cm long was attached to the low point of the trough. The cribs were held in place and separated at 11 cm intervals by wooden separators clinch-nailed in place to the sides of the module.

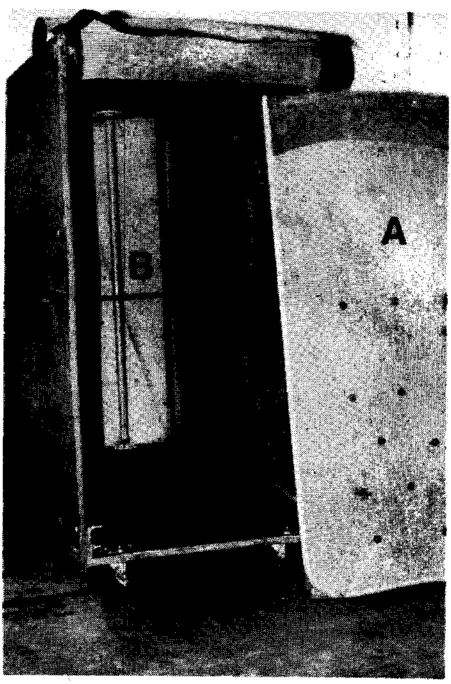

FIG. 1. Wheat-holding crib and Sitotroga cerealella production module (horizontal position). A. Crib. B. Module in horizontal position showing collection trough and steel pipe crib supports.

Two 2.5 cm diam. steel pipes ca. 1.28 m long were threaded to accept flanges at each end. The flanges were then bolted to the ends of the module ca. 25 cm apart so that they ran longitudinally just above the top edge of the collection trough. These pipes held the cribs at a uniform level and also strengthened the module. A 7.6X102 cm ventilation port covered by nylon screen (7.9 mesh/cm) was located on each side of the module at the level of the steel pipe supports. Four 7.4 cm heavy-duty casters were bolted in place at each corner of the end of the module opposite the collection tube of the trough so the crib-loaded module could be easily moved when in a horizontal position. A 61X122 cm steel frame constructed of 2.5 cm angle iron was fitted to the bottom of the module. This frame had 7.4 cm heavy-duty casters attached at each corner so the crib-loaded module could be easily moved when in a vertical position. The frame was attached by 10X10X0.7 cm steel plates welded to the sides of the frame which, when drilled with 0.7 cm holes, allowed the frame to be bolted to the module. A heavy-duty 2.5 cm diam. eye bolt was attached to the module 10 cm down from the top center of the end of the module with the collection tube. It was bolted from the inside with a 5 cm washer so the unit could be lifted from the horizontal position to the vertical with the aid of a ceiling- mounted winch. The top inside corners of the module were also reinforced with 7.5X7.5 cm steel corner braces bolted in place at each corner.

Infestation and Immature Development. After the 10 grain-filled cribs placed in the module (Fig. 2), the grain was infested with enough viable *S. cerealella* eggs (24-48 h old) to yield ca. two larvae/kernel of wheat. This required 100g of *S. cerealella* eggs (50,000 eggs/g).

Before infestation, the eggs were decontaminated in a 10% formalin solution for 10 min, rinsed in fresh water for 5 min, and then placed in 5 L of water. This egg solution was distributed uniformly over the top of the horizontal positioned cribs with a simple infestation wand made of galvanized pipe (0.6 cm inside diam.) joined to form a "T" (Fig. 2-A). The vertical pipe of the "T" was ca. 150 cm and the horizontal pipe was ca. 50 cm. Holes of 0.2 cm diam. were drilled at ca. 1.7 cm intervals along the horizontal pipe. In operation, the egg-water solution was poured into an appropriately-sized vessel equipped with a bottom spout. The vessel was then placed on a magnetic stirrer (which kept eggs in solution) at a level above the highest crib. A 3 m length of 1.7 c diam. clear plastic tubing connected the vessel and the wand. The wand was uniformly passed over the top length of each crib until ca. 500 ml of solution/crib was dispersed. The operator regulated the flow of the solution by crimping the plastic tube.

During infestation, small amounts (4-5ml) of the egg solution were placed on paper towel. This towel was dated and reserved to determine egg viability by counting the hatched and unhatched eggs (ca. 300 total) on a randomly selected strip of towel after 6-7 days. Egg viability ranged between 85-95%.

After infestation, the module containing the infested cribs was kept in the horizontal position for 12 days for egg hatch and larval penetration of the grain. On day 13 postinfestation, the module was brought into the vertical position and a thermometer probe was placed into the middle of one of the center cribs to monitor grain temperature. On day 15 postinfestation , a nylon screen (7.9 mesh/cm) was used to seal the open top of the module. The screen was attached to the module with Velcro®, a two-piece nylon self-closing device (Velcro Inc. New York, N.Y.) (Fig. 2-B). After this, a ventilation fan was placed on top of the unit to dissipate the excessive metabolic heat generated by the developing larvae at the high-level grain infestation (ca. 80%). The fan was made from two-1/20 HP, 1550 RPM electric motors driving 38 cm diam. fan blades. The fans had wire cage guards which were bolted in tandem to two 130 cm lengths of 2 cm electrical conduit tubing spaced 30 cm apart. The tubes rested on top of

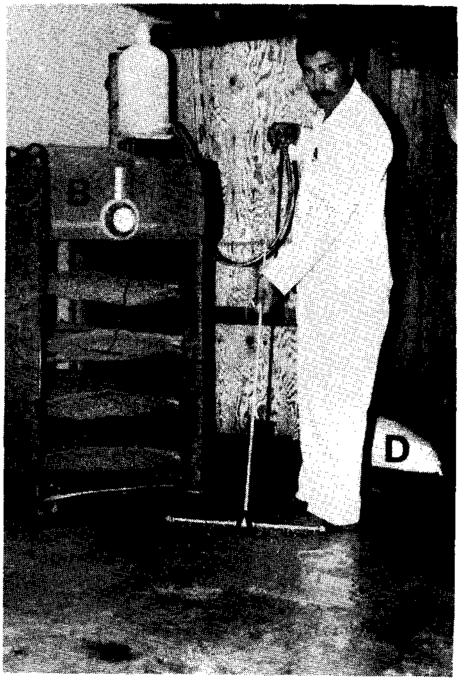

Fig. 2. Sitotroga cerealella egg application device and horizontal and vertical production modules. A. Egg application device. B. Velcro-attached top screen. C. Ventilation fan unit. D. Adult collection bag.

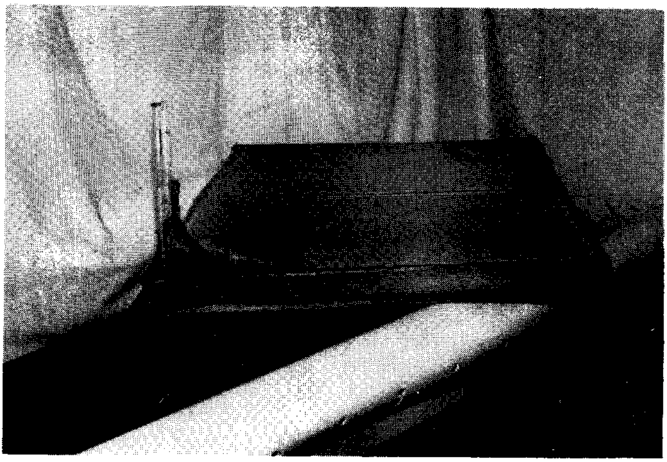

FIG. 3. Sitotroga cerealella oviposition frame on galvanized metal pan.

the module which supported the ventilation unit off the screen (Fig. 2-C). The fans were necessary since estivation is induced in immature stages of S. cerealella if they are exposed to temperatures above 35°C (Morrison and Hoffman, 1976). The thin layer of grain combined with constantly moving ambient air (26.7°C) over the cribs was sufficient to keep grain temperature below 32°C and maintained a near desirable temperature for immature development (Boldt 1974).

Adult Production and Collection. First emergence of adults began ca. day 22 postinfestation and collections were made each 24 h thereafter until day 40 postinfestation. After the final collection, the module was immediately placed at 66°C for 24 h. Then the cribs were emptied, the spent grain disposed, the module cleaned, and the unit returned to service.

The adults were collected by placing a 40X50 cm cloth muslin bag over collecting cone of the module (Fig. 2-D). A 5 cm length of polyvinyl chloride plastic pipe with an inside diameter of ca. 8 cm was glued to one corner of the bag. The pipe was a slip-fit on the collecting cone of the module. After bag attachment, the sides of the module were gently struck with a rubber mallet to dislodge adults from the sides of the cribs and the module. An air gun operated at ca. 3.5 kg/cm^2 was then used to first blow the adults down onto the collection trough, then a 3 cm hole previously drilled in the end away from collection cone allowed access of the air gun to blow the adults off the trough into the collection bag. A cork was used to close the 3 cm hole when not in use. After collection, the bag of adults was removed and the collection cone closed. The adult weight was determined and then they were dispensed into oviposition frames.

Oviposition, Egg Collection and Egg Cleaning. The oviposition frame (90X70X3.8 cm) was made of 3.8X2.5 cm smooth Douglas fir. A center piece divided the frame into two 45X70 cm units. The frame was covered on one side with 7.6 strand/cm nylon screen. The other side was covered with 13 strand/cm plastic screen. Previous work (Morrison and Hoffman 1976) showed the 7.6 strand/cm screen elicited high rates of oviposition; the 13 strand/cm screen contained the adults yet allowed good ventilation. One outside corner of each of the 45X70 cm units had a 30 cm piece of 2 cm wide Velcro "hooks" sewn to the 13 strand/cm screen and the matching "eye" Velcro stapled to the wood frame (Fig. 3). This formed an easily operated, insect-proof device to load and empty the frame. Previous work had shown that ca. 500 g of adults (125,000 individuals) dispensed into each unit of the frame was a favorable ratio of adults/cm^2 for oviposition (Morrision and Hoffman, 1976). When loaded, the frame was placed (7.6 strand/cm side down) on a 100x75 cm galvanized sheet metal pan (26 gauge) that had first been lightly dusted with "Dry-Flo", an industrial starch powder (National Starch and Chem. Corp. Bridgewater, NJ 08807). The frame and pan were then placed as a unit on a horizontal storage rack for 72 h. During this period, the females deposited eggs through the screen onto the pan, on the screen, and in clusters within the frame. Daily during the 72 h oviposition period, the eggs were collected by taking the pan and frame to a large working table where the frame was removed from the pan and turned upside down. The 7.6 strand/cm screen was then vigorously brushed with a 7.5 cm paint brush to dislodge eggs adhering to the screen. After brushing, the frame was then turned over and vigorously shaken over the metal pan for ca. 30 sec. so that eggs within the frame would fall out. After this, the metal pan was brushed free of the eggs and Dry-Flo, redusted with Dry-Flo, the frame replaced, and the unit replaced on the rack. Those frames that had been processed three times were washed and dried after discarding the moths.

The mass of eggs, Dry-Flo and insect parts was collected and placed in a standard sieve series with sieve nos. 20, 30, 40, 50, 60 and a catch pan which had been placed on an automatic sieve shaker. The shaker was then

TABLE 1. Mean Daily Production of *Sitotroga cerealella* Eggs and Return of Eggs from Initial Adult Weight from a 12-wk Period for three 3 Consecutive Yrs.[a/]

Wk beginning	1981 Eggs (g)	1981 Wt. return (%)	1982 Eggs (g)	1982 Wt. return (%)	1983 Eggs (g)	1983 Wt. return (%)
June 15	546.9	18.6	546.6	19.4	309.3	18.1
22	568.6	20.3	408.3	18.5	442.6	16.2
29	599.1	20.6	544.1	17.5	543.2	19.6
July 6	542.8	19.1	564.1	17.0	441.3	19.9
13	543.0	20.8	315.5	12.9	655.3	19.7
20	496.1	20.5	214.1	10.4	650.0	19.2
27	490.4	19.1	392.4	13.4	584.0	17.5
Aug. 3	573.0	22.7	441.3	15.5	302.5	18.2
10	420.0	17.8	565.8	15.9	354.8	15.6
17	497.3	20.3	460.4	17.1	755.1	20.3
24	321.7	19.9	343.8	16.0	475.1	18.6
31	273.4	19.3	417.9	15.2	432.0	20.2
$\overline{X}$	489.4	19.9	434.5	15.7	495.4	18.6

[a/] 39 kg Wheat infested/day.

TABLE 2. Cost Analysis for Eggs of *Sitotroga cerealella*.

Item	Cost	Daily cost
184 m^2 building space @ \$543.00/m^2	\$99,912.00	
Depreciate 20 yrs (7300 days)		\$13.69
44 production modules @ \$1000.00 ea.	\$44,000.00	
25 oviposition frames @ \$30.00 ea.	750.00	
Other equipment	\$25,000.00	
	\$69,750.00	
Depreciate 10 yrs (3650 days)		19.11
Wheat - 91 kg @ \$.27/kg		24.57
Labor - 19 h/day @ \$10.00/h		190.00
Utilities @ \$50.00/day		50.00
Maintenance @ \$10,000 yr - \$27.40/day		27.40
		\$324.77

Egg cost $\frac{\$324.77}{49{,}600{,}0000 \text{ eggs/day}^{a/}}$ \$.00655/1000

[a/] Derived from 91 kg wheat infested/day x eggs/kg less 600,000 eggs/day reserved for reinfestation.

operated for ca. 5 min. After this, insect parts in the #20 sieve and the Dry-Flo in the catch pan were discarded; egg clusters on the #30 sieve were held separately and used for grain infestation. Eggs that remained in the other sieves were combined and poured through a horizontally moving air current generated by a simple wall-mounted, variable speed ventilation fan until the very fine insect scales not separated by the sieves were removed. The eggs were then placed in dated, open-top containers and stored at 13°C.

A temperature of 26°±2°C and a relative humidity of 75±5% were maintained in all rearing areas and lights were used only when rearing areas were occupied by working personnel. Each production module required ca. 3 m^2 of working space and the oviposition room was 8 m^2. For protection against insect scales, all personnel wore coveralls, protective dust masks, and/or Whitecap® positive pressure breathing hoods (3M Co. Inc., St. Paul, MN 55101) when working in the rearing areas.

RESULTS AND DISCUSSION

During the 3 yrs of production reported in this paper, 39 kg of wheat/day were infested from May through August of each year. During the remaining 8 months of each yr, only 13 kg of wheat/day were infested.

Data in Table 1 show that over a 3 yr period, large numbers of *S. cerealla* eggs were produced during the peak 12 wk period of demand ($\bar{x}$ = 473.1 g/day). The weight return of eggs from initial collected adult weight was 18.1%, a ratio of ca. 600,000 eggs/kg of infested wheat.

Maintence of the colony of *S. cerealella* at the infestation rate of 39 kg of wheat/day required ca. 8 man h/day on a 7 day/wk basis.

Cost of wheat over the 3-yr period was ca. $.27/kg but this could be reduced by ca. 1/2 or more if purchased by contract.

Although not shown, production records over the last 11 yrs show the return of ca. 600,000 eggs/kg wheat infested was achieved with infestion rates that ranged from 91 to 6.5 kg wheat/day. This system is therefore ca. 60% efficient when compared to a theoretical 100% efficient system where all wheat kernels were infested and produced adults (based on 28,000 kernels of wheat/kg, 250 adults/g with a 1:1 sex ratio and 60 eggs/female). A cursory cost analysis based on a continous infestation of 91 kg wheat/day and an 18.1 % weight return of eggs from adult weight produced show the cost of eggs to be $.00655/1000 (Table 2).

LITERATURE CITED

Boldt, P. E. 1974. Effects of temperature and humidity on development and oviposition of *Sitotroga cerealella* (Lepidoptera:Gelechiidae). J. Kan. Entomol. Soc. 47:30-36.

Flanders, S. E. 1930. Mass production of egg parasites of the genus *Trichogramma*. Hilgardia 4:465-501.

King, E. G., R. K. Morrison, and F. R. Ferrer. 1978. Production of tachinids, trichogrammatids, and their hosts for control of arthropod pests by augmentation with emphasis on lepidopteran pests of sugarcane. Proc. Seminario Nacional Sobre el Problema de los Taladradores de la Cana de Azucar. pp. 95-122.

Morrison, R. K. and J. D. Hoffman. 1976. An improved method for rearing the Angoumois grain moth. USDA, ARS-5-104. 5 pp.

Morrison, R. K. and E. G. King. 1977. Mass production of natural enemies, pp. 173-217 *In* R. L. Ridgway and S. B. Vinson, eds. Biological Control by Augmentation of Natural Enemies. Plenum Press, New York.

Spencer, H., L. Brown, and A. M. Phillips. 1935. New equipment for obtaining host material for mass production of *Trichogramma minutum*, an egg parasite of various insect pests. U. S. Dept. Agric. Circ. 376, 17 p.

Wilkinson, J. D. and R. K. Morrison. 1972. Effect of grain treatment and infestation level on larval infestation and adult emergence of the Angoumois grain moth, Sitotroga cerealella, reared in wheat. J. Econ. Entomol. 65:264-268.