Synthesis Report on Rearing Asian Longhorned Beetle

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Abstract

Since not all research on *Anoplophora glabripennis* (Motschulsky) (ALB) can be conducted in China or at North American sites where it is being eradicated, the ability to mass rear the Asian longhorned beetle is critical to rapid progress on research necessary for exclusion, detection, and eradication of this serious pest. Large numbers of beetles in all life stages are needed year round. Currently, research on rearing methods is being conducted at three primary rearing sites: Cornell University, Ithaca, NY; USDA Forest Service, Quarantine Laboratory, Ansonia, CT; and USDA Animal and Plant Health Inspection Service (APHIS) Otis Plant Protection Center, Otis Air National Guard Base (ANGB), MA. The advantages and disadvantages of the modified diets currently in use are summarized in Table 1. Current methods and diets used at the three sites are summarized in Table 2.

ALB can complete its development on all three diets in use; these are modifications of previously published diets. The formulations in use are the result of studies that compared available diets or modifications to them. Studies at Ansonia showed that use of the modified red oak borer diet reduces handling and subsequent contamination, shortens developmental time, and results in pupation that exceeds 60 percent. These studies also showed that higher levels of available iron in the diet slow larval development, increase larval mortality, and reduce percent pupation.
Larval growth and adult parameters were compared at Ithaca using artificial diets developed for three species of *Lamiini*, including one diet developed for ALB in China. The only difference in performance of larvae and adults reared on these three diet types was that nondiapausing larvae reared on *Monochamus carolinensis* (Oliver) diet (Necibi and Linit 1997) requireed less time to pupate than larvae reared on ALB diet. Comparisons of diets with cellulose and wood products, also studied at Ithaca, showed that males grew fastest on diets with sawdust or phloem-cambium and remained for the shortest period as pupae on ALB diet. Females grew faster on diets with cellulose than sawdust and lived the longest as adults on the ALB diet.

Mass rearing of ALB requires not only on a good artificial diet but one that can be dispensed into many containers relatively quickly. A twin-screw extruder is being installed at the APHIS quarantine laboratory that can dispense diets with a wider range of physical characteristics than can be accommodated with diets that must be "pourable" before fully solidifying. Diets dispensed by this machine will be tested as part of an effort to develop systems that provide appropriate larval nutrition and environment but are more resistant to degradation and can be dispensed easily.

A great concern in laboratory rearing is that the individuals reared on artificial diet may not be comparable to those reared on host material. Adults from larvae reared on the modified red oak borer diet at Ansonia have been used for both flight and oviposition studies, and performed as well or better than adults that emerged from infested wood.

We have also developed methods for holding and handling each life stage of ALB. Eggs can be allowed to hatch under the bark of oviposition bolts, though this is not always desirable because bolts can dry out quickly. This increases egg mortality, larval mortality during establishment, microbial contamination of the diet when the larvae are taken from wood, and asynchrony of larval development. As a result, methods for collecting and in vitro hatch of ALB eggs have been developed and are being used at both the Forest Service and APHIS quarantines. When larval rearing temperatures were compared at Ansonia, mortality over the first 8 weeks was higher at 20°C than at 25°C despite lower microbial contamination. Development at 25°C was more rapid than at 20°C, but there was the potential for heavier pupae at 20°C than at 25°C. Methods for retrieving and holding prepupae and pupae

Table 1.—Advantages and disadvantages of the three modified diets currently in use

Item	Modified ALB diet	Highly modified red oak borer diet	Modified tilehorned *Prionus* diet
Advantages	1) Resists microbial contamination well 2) Less frequent diet changes required 3) Fairly rapid development 4) No host material	1) No special mixing required; can be poured directly into containers 2) Fairly rapid development 3) No host material	1) No special mixing required; can be poured directly into containers 2) No host material
Disadvantages	1) Must be hand mixed and packed into containers 2) Contains ingredient that is irritating to humans	1) Diet must be changed frequently 2) Can develop microbial contamination	1) Diet must be changed frequently 2) Can develop microbial contamination 3) Fairly slow development

Table 2.—Current methods and diets used at three quarantine rearing sites

Item	Cornell University Ithaca, NY	USDA Forest Service Ansonia, CT	USDA APHIS Otis ANGB, MA
Diet in use	Modified ALB diet (Diet D, Zhao et al. 1999)	Highly modified red oak borer diet (Galford 1985)	Modified tilehorned *Prionus* diet (Diet 3; Payne et al. 1975) and modified ALB diet
Rearing temperature	22.5 ± 2.5°C	25 ± 2°C or 20 ± 2°C	23 ± 2°C
Rearing relative humidity	Larvae, pupae, and logs > 75% RH, Adults 60% RH	First 2 weeks ~100% RH; then 60 ± 5% RH	First 4 weeks ~100% RH; then 55 ± 15% RH
Larval rearing containers	59-ml plastic cups 237-ml glass jars	59-ml, 118-ml, and 237-ml plastic jars	50 ml wells in plastic trays and 237 ml glass jars
Frequency of diet changes	First change at 12 weeks, all subsequent changes at 4 weeks; diet change just prior to chill is the last	Every 2 weeks at 25°C and every 3 weeks at 20°C	Every 2 weeks (first 2 changes), then every 3 weeks
Larval chill	At 180 days chill at 5°C for 30 days	At ~100 days chill at 10°C for 112 days	None
Adults containers	926-ml (singles) or 3.9- liter (pairs) glass jars	950-ml (singles) or 3.8-liter (pairs) glass jars	2-liter clear, vinyl jars (singles) or screen cages (pairs)
Host used for adult feeding and oviposition	*Acer saccharum*	*Acer saccharum*	*Acer* spp., usually *Acer pensylvanicum*

developed at Ansonia greatly reduced eclosion abnormalities that occur when larvae pupated in the diet or were exposed to low humidity. Additionally, methods for holding adults in use at Ithaca and Ansonia have increased survival time and curtail aggressive behavior. Because some late instar larvae stopped developing and did not resume development until after a cold period, research continues at both Ithaca and Ansonia to determine ideal conditions and durations of chill.

Some research requires bioassaying even-aged and even-sized larvae, so methods to synchronize development are being sought. Methods for stockpiling eggs by chilling oviposition bolts and synchronizing egg hatch have been developed at Ansonia. These methods provide even-aged larvae and lengthen the time that small larvae are available. A chill period during the later larval instars also has been shown to shorten the time to pupation and improve the synchrony of adult eclosion.

It would be beneficial to have adults available for research year round and eliminate the need for using fresh host material for feeding and oviposition. Prototype artificial logs for oviposition have been developed at the APHIS quarantine. Logistics for making and using the logs, along with their acceptance by female beetles, is less than ideal, so additional work in this area is needed.

Because the supply of ALB for research is limited and there are few quarantine laboratories with permits for this species, researchers have sought a surrogate species that need not be quarantined. The cottonwood borer, *Plectrodera scalatoa*, a closely related native cerambycid, was chosen for this purpose. Leah Bauer and Debbie Miller (USDA Forest Service, East Lansing, MI) developed a method for rearing the cottonwood borer in the laboratory using the tilehorned *Prionus* diet.

Much progress has been made toward maintaining a readily available supply of healthy ALB for research purposes. However, to mass rear ALB, we need to: 1) Reduce the time and cost of rearing individuals from egg to adult; 2) Increase the resistance of some diets currently in use to microbial contamination; 3) Better understand the requirements for large larvae to initiate pupation; 4) Increase quarantine space and resources to rear larger numbers of beetles at some sites; 5) Develop an artificial diet and oviposition substrate for adults or methods to retain host material over the winter so that all stages are available year round; and 6) develop a method for rearing larvae on cut host material or for obtaining naturally reared individuals for studies that require them.

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