Eucalyptus killed by eucalyptus borers.

Introduction and Establishment of Natural Enemies of the Eucalyptus Longhorned Borer

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Eucalyptus are among the most abundant ornamental, shade, and windrow trees in California. They have long been valued for their fast growth, and tolerance of drought and marginal soil conditions (FAO 1981). Eucalyptus were also free of significant insect pests until the eucalyptus longhorned borer (ELB) appeared in Orange County in 1984 (Scriven et al. 1986). Larvae of this Australian beetle destroy the cambium of eucalyptus trees, usually killing the trees in a matter of weeks (Hanks et al. 1991, 1993a, Paine et al. 1995). ELB quickly spread throughout southern California, where it has killed tens of thousands of trees. The beetle continues to move northwards, recently appearing in Santa Barbara, Kern, Fresno, Sacramento, and Glenn Counties, and it is now spreading along the east side of San Francisco Bay. Continued loss of trees will degrade California landscapes substantially. Furthermore, removal and replacement of dead trees will cost millions of dollars per year for the foreseeable future.

In 1995, we identified the beetle *Phoracantha recurva*, in the same genus as the eucalyptus longhorned borer (*Phoracantha semipunctata*), for the first time in California. *P. recurva* is similar in appearance to ELB, but differs in its biology, attacking the upper branches of eucalyptus while ELB prefers the trunk. Colleagues in South Africa report that the two species work in tandem, delivering a one-two punch to eucalyptus trees (G. Tribe, pers. comm.). This new beetle is expected to quickly increase its numbers and distribution.

At this point, biological control with natural enemies appears to be the only feasible option for controlling these pests and limiting further tree kills, and we have laid the groundwork for a biological control program for these pest species. We identified five parasitic wasp species (an egg parasite and four larval parasites) which attack the eggs and larvae of the beetles (Austin et al. 1994). These parasites have been imported into California from Australia and established in culture (Hanks et al. 1995a). Mass rearing protocols have been worked out and optimized for each species. We have reared and released some 200,000 parasites of *Phoracantha* since 1993, in San Diego, Orange, Riverside, Los Angeles, Fresno, Santa Barbara, and Santa Clara counties. To date, the egg parasite is firmly established in southern California, where it is finding and killing 90% of the ELB eggs (Hanks et al. 1996, in press). We also have evidence that one of the larval parasites is established at release sites in Riverside and Santa Barbara counties. In this final report, we summarize our progress in 1995, and for the project overall.

Project Objectives:

1. To establish natural enemies of the longhorned borers *Phoracantha semipunctata* and *P. recurva* in California.

2. To determine the effectiveness of natural enemies of the *Phoracantha* borers by assessment of parameters such as the percent of pest insects killed by the introduced natural enemies, decline in population density of the pest species, and decline in tree damage and mortality.

Eucalyptus borers *P. semipunctata* (left) and *P. recurva* (right).

Parasitoid *S. lepidus* parasitizing eucalyptus borer larva.

Methods and Materials

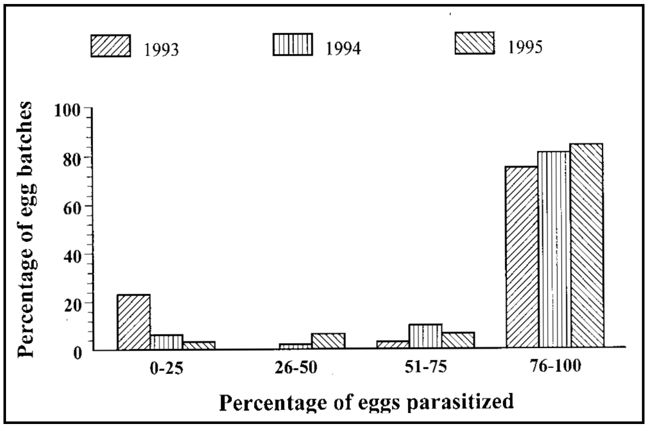
Mass Rearing of Parasites. We selected five species of parasites for introduction, based on consideration of the most common species found parasitizing *Phoracantha* beetles in their home ranges in Australia. These include the larval parasites *Syngaster lepidus*, *Callibracon limbatus*, *Jarra maculipennis*, and *J. phoracantha*, and the egg parasite *Avetianella longoi*. Both *Phoracantha* species are readily attacked by all five of the parasite species which we have imported and reared. All parasite species can only be reared on eggs or beetle larvae, so we must maintain a large and continuous colony of ELB on eucalyptus logs. Briefly, fresh logs are infested with ELB larvae, and the larvae are allowed to develop to the size optimal for a specific larval parasite species. Most of the logs are then exposed to one of the larval parasite species for 1 week, then moved sequentially to rearing and emergence cages. The emerging adult parasites are collected and released in batches at field release sites. Production of the larval parasites is limited only by the number of ELB-infested logs we can generate and process (e.g., in 1994 and 1995, approx. 4,000 logs were processed annually).

The remainder of the logs are kept until the adult ELB beetles emerge. The adults are transferred to oviposition cages, where the females lay egg masses on filter paper sheets. Most of the ELB eggs are used immediately as hosts for the egg parasite, with the remainder being allowed to hatch, and the developing larvae transferred to fresh logs to maintain the ELB colony.

The egg parasites (*Avetianella longoi*) are reared in plastic shell vials provided with freshly-laid ELB egg masses on filter papers. Just before the adult parasites emerge, the parasitized egg masses are transferred to logs at release sites, as described below. Production of egg parasites is limited only by the number of host ELB eggs which we can harvest from the lab colony.

Release of Parasites. Release sites are chosen on the basis of two criteria. First, they must have significant ELB infestations, to provide abundant host material, and give the parasites the best possible chance of establishing. Second, they must be secure, to prevent theft of parasite-containing logs for firewood, or vandalism.

Release sites are provided with piles of fresh-cut eucalyptus logs to encourage natural ELB oviposition, with further fresh logs added weekly. We have successfully established parasites as follows: Fresh-cut eucalyptus logs are piled on a platform to attract ELB adults, and eggs deposited by these beetles serve as hosts for the egg parasite, and any surviving larvae are hosts for the larval parasites. Log piles are set up several weeks prior to release of parasites, and at least one new log is added weekly to ensure a continuous supply of parasite hosts. The egg parasite, *A. longoi*, is released by placing parasitized egg masses from the laboratory colony under loose bark flakes on the piled logs. Emerging parasites can therefore imprint on eucalyptus log odors/characteristics, and host eggs deposited by ELB are available. The larval parasites are released by collecting freshly-emerged adults from the laboratory



Parasitism for longhorned borer egg batches collected in the field in San Diego, Rancho Santa Fe and Riverside during Summer, 1993. (Fig. 1)

colonies, transporting them in screen cages with water and honey provided, and freeing them at the log pile. Released larval parasites often parasitize borer larvae in the log pile before dispersing, their progeny emerging as adults about one month later.

Parasites are released in large batches (several hundred to several thousand, depending on species) at a limited number of selected sites to ensure that a given site receives the critical mass of parasites required for establishment.

Monitoring Establishment and Efficacy of Parasites. Egg parasite establishment is monitored first by collecting naturally-laid *Phoracantha* egg masses on trap logs set out at release sites, and both visually determining whether any eggs are parasitized, and subsequently collecting the eggs to capture and count the parasites as they emerge. Once parasitized eggs are found, systematic surveys for *Phoracantha* egg masses on fallen branches and logs are carried out, at increasing distances from the release point. Egg masses are evaluated for the percent parasitized (i.e., found by the parasites) and the percent parasitism within an egg mass, to assess the overall efficiency of the parasites.

Establishment of larval parasites is determined by analogous searches for parasite cocoons and/or emergence holes in trap logs, followed by surveys of natural windfalls and fallen branches. Parasite efficiency will be assessed once populations are well established by determining the percentage of *Phoracantha* larvae which are parasitized, as shown by parasite cocoons and emergence holes, versus the number which escape parasitism, as determined by the number of (much larger and different shaped) *Phoracantha* emergence holes.

Results

Optimization of Parasite Mass Rearing. The solitary larval parasites *S. lepidus* and *C. limbatus* had similar life histories in culture, with egg-adult development taking about one month. Each species exhibited strong host-size preferences, with the smaller *S. lepidus* accepting smaller hosts. Furthermore, discrimination occurred within these overall size classes, with hosts at the small end of the preferred host range scale for each species producing exclusively male progeny, medium-sized hosts producing mixed males and females, and large hosts producing exclusively females (the larger sex in both species). In both species, males generally emerged before females, and females were mated immediately upon emergence by waiting males. The duration of mating was very brief, lasting only a few seconds.

The gregarious parasites *J. maculipennis* and *J. phoracantha* (egg-adult also ~4 weeks) showed no distinct host size preferences. Instead, clutch size was adjusted according to host size. The sex ratio was strongly female biased, with only 1-2 males produced per clutch.

Releases of Parasites and Monitoring Establishment. We released *A. longoi* at the following sites in California during 1993: Rancho Santa Fe and UC San Diego (both in San Diego Co.), UC Riverside and the city of Riverside (Riverside Co.), Will Rogers State Park (Los Angeles Co.), and Stanford University (Santa Clara Co.). The parasite established at 4 of the 6 sites, and because the wasps were widely distributed and abundant at these sites, there was no need for further releases. Releases of *A. longoi* in 1994 were restricted to areas where they had not established (Stanford, Will Rogers State Park) or had not yet been released (Irvine, Long Beach, Santa Clarita, Santa Barbara). In 1995, we added release sites in Fresno (Fresno Co.), and Chollas Lake Park (San Diego Co.). During summer of 1996, we will be checking for establishment at all of these sites, and beginning releases at new sites (several sites in the San Francisco Bay area which are now under heavy borer attack, as well as a site near Corning).

Because *A. longoi* only require ELB eggs for development, they are relatively easy to rear in large numbers and we were able to release more than 53,000 *A. longoi* in 1993, 65,000 in 1994, and 56,000 in 1995.

However, the larval parasites require three-week-old host larvae in eucalyptus logs and so are much more difficult and expensive to produce. We reared and released 1,000 *S. lepidus* at the Rancho Santa Fe, U.C. San Diego, U.C. Riverside, Will Rogers State Park, and Stanford University sites in 1993. Improvements in our rearing methods enabled us to release about 14,000 wasps in 1994 and 1995 at sites in Fresno, Chollas Lake Park, Stanford, Rancho Santa Fe and Santa Barbara. Evidence for breeding populations of *S. lepidus* has been found at two release sites (Santa Barbara and U.C. Riverside) with observations of adult emergence holes and of ovipositing adults in the field. Intensive monitoring of sites will continue during summer 1996 to confirm establishment.

Efficacy of the Egg Parasite, *A. longoi*. The egg parasite is firmly established and spreading rapidly throughout southern California. For example, well established parasite populations were found in Hemet in 1995, about 30 miles from the nearest release sites in Riverside. *A. longoi* shows great promise as a biological control agent by effectively locating ELB eggs in urban and rural habitats. Figure 1 summarizes percentage parasitism data for longhorned borer egg batches collected in the field in San Diego, Rancho Santa Fe, and Riverside during Summer, 1993 (N = 40 batches), 1994 (N = 97 batches), and 1995 (N = 63 batches). More than 75% of the egg batches collected in 1993 had over 75% of their eggs parasitized. In Summer, 1994, an even greater percentage of egg batches were over 75% parasitized, even though we had not released parasites at these sites since the previous year. Eggs collected in 1995 showed the same pattern, with more than 80% of batches showing high parasitism rates. For all three years, more than 90% of eggs collected from the field were parasitized and killed by *A. longoi*. Clearly, this parasite is a highly effective natural enemy of ELB in California and will prove valuable in reducing the economic impact of the beetle.

Communication/Outreach

Our program has involved a large network of cooperators, including arborists, grounds keepers, parks officials, and other landscape professionals, who are active participants in locating infestations and release sites, and assisting with parasite releases and monitoring of release sites for parasite establishment. Up to date information has been disseminated through articles in the popular press, newsletters, fact sheets, and technical and academic journals. We have also given presentations and workshops at the local, state, and national level, and we handle a large number of calls from homeowners, farm and pest control advisers, and public officials.

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