Susceptibility of the eastern tent Caterpillar (Malacosoma americanum) to the Entomogenous Fungus Beauveria bassiana

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Falacosoma americanum (Fabricius) (Lepidoptera L: Lasiocampidae), the Eastern tent caterpillar, is a pest native to the United states. It is particularly injurious to apple, cherry, and other fruit trees, but can be destructive to other deciduous trees. Control of this pest is difficult, largely because of its congregation behavior. Larvae spend the day inside of their protective tents, where they are largely shielded from exposure to insecticides. At night and in rainy weather they emerge to forage, at which time a widely disseminated control would be necessary. Malathion and Bacillus theriengensis preparations have been used to control M.americanun, although diurnal cutting and removal of individual tents is often the recommended treatment. Efforts to develop new biocontrol agents against M. americanum have focused on parasitic wasps and entomogenous nematodes (Nielsen, 1989). However, because of the insect's congregative lifestyle, If even a few larvae are successfully infected by a contagious agent during a foray, they may return to the tent to die and eventually infect the entire colony.

The entomopathopathogenic fungus *Beauveria* bassiana is widely regarded as one of the most promising species known for potential development into a practical insect biocontrol agent. Unlike bacterial, viral, or protozoanent mopathogens, fungi need not be consumed by their hosts in order to be infective. Instead, germinating fungal spores are able to grow directly through the insect's cuticle. It has been suspected that entomopathogenic fungi are enabled in this novel mode of infection by the production of cuticle-degrading enzymes (Smith *et al.*, 1981). Larvae of *the Eastern tent caterpillar. (M. americanum*) were collected locally and maintained on a diet or fresh leaves from the wild choke cherry (*Prunus anginiana*).

Healthy larvae of M. americanum (approximately

0.1 g/larva) were placed in plastic dishes in groups of 10 and suspension containing 6 x 107 spores in water (i.e., 7.2×10^7 spores ml). Treated larvae and water controls were then maintained on their diet and scored daily for mortality.Liquid cultures of B. bassiana strains were grown on a basal salts medium (0.06% MgSo₄, 0.05% NaCl, 1.5% KH₂PO, 0.001% FeSO₄, 7H₂O, 0.001% ZnSO₄) containing 0.5% (w/v) insect cuticle. Cuticles from larvae of *M. americanum* were prepared by dissection. Media containing cuticle were subjected to heating at 65°C for 15 min to inactivate any endogenous enzymes. Cultures were inoculated to a concentration of 2, 10 spores/ml and incubated on a shaker at 200 rpm 26°C for 96 hr. Supernatants were prepared by centrifugation at 2000g for 10 min and stored at 20°C for enzyme assays. In preliminary studies, we found no effect of dialyzing samples, so this step was omitted.

Assays of cuticle-degrading enzymes were carried out as previously described (Gupta *et al.*, 1991). One endochitinase unit is defined as the change of one optical density at 540nm per 45hr of incubation at 37°C. Other enzymes are expressed in international units. Protein was determined by the procedure of Bradford, 1976. Using bovine serum albumin as a standard protein.

Since preliminary experiments indicated that enzyme levels were both too low to be measured in early growth cultures and relatively stable in stationery phase (96 hr). Since the insolubility of cuticle made dry weight measurements impractical, activity values were normalized by total extra cellular protein. Enzyme values are presented as the mean and standard deviation of three or more measurements and are expressed in units per milligram extra cellular protein rounded to three significant figures.

"Larvae of *M. americanum* were inoculated with suspensions containing 7.2×10 spores ml. Treated larvae