Invasion Genetics of Emerald Ash Borer (*Agrilus planipennis* FAIRMAIRE) in North America

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Emerald ash borer (EAB) was first detected in Michigan and Canada in 2002. Efforts by federal and state regulatory agencies to control this destructive pest have been challenged by the biology of the pest and the speed in which it has spread. Invasion dynamics of the beetle and identifying source populations from Asia may help identify geographic localities of potential biocontrol agents. Genetic techniques, such as mtDNA gene sequencing, amplified fragment length polymorphisms (AFLP), nuclear gene sequencing, and microsatellite analysis will help determine the geographic origin of the native east Asian population(s) of EAB that have given rise to the EAB that is invasive in North America.

In North America, EAB has been detected in much of the Lower Peninsula of Michigan and Ontario, Canada, as well as northern Indiana and Ohio. To represent the diversity of the introduced population, we have obtained approximately 2,100 EAB individuals, mainly from 32 localities in Michigan, but also from three localities in Ohio and one locality each in Indiana and Ontario. DNA was isolated from EAB individuals from seven localities in China, four localities in South Korea (kindly loaned to us by Dr. Dave Williams, USDA-APHIS), and one adult from Shiroishi, Japan (kindly loaned to us by Dr. Paul Schaefer, USDA-ARS) to represent the native range of EAB in Asia. Identical mitochondrial cytochrome oxidase I (COI) sequences (485 bp) were obtained from all North American EAB analyzed (Michigan [76], Ohio [2], Indiana [4], and Canada [6]), all EAB from China (Dagong [5], Hangu [1], Heilongjiang Province [4], Hebei [2], Jilin [2], and Liaonging [2]), and six EAB individuals from three localities in South Korea. However, mitochondrial COI sequences from five individuals in two populations in South Korea differed from this common haplotype by two to four nucleotides, and the Japanese sample differed from the common EAB mtDNA haplotype by 3.7 percent. Therefore, the mtDNA COI sequence of the Japanese sample is very different from any other individual sampled and there is COI haplotype variation in two of the three localities in South Korea. We have obtained AFLP profiles from EAB individuals from Michigan (46), Ohio (2), Ontario(6), South Korea (4), Dagong (4), Heilongjiang Province (3), Liaonging (2), Hebei (2), Jilin (2) and 1 individual from Japan (four selective AFLP primer pairs; 139 scoreable loci). We have observed differences in AFLP profiles, both within populations from the same location, as well as between all

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populations. Neighbor-joining analysis of the 139-band AFLP data set indicates that individuals from MI cluster more often with individuals from China, while the Japan individual fell into a separate, more distantly related group. However, we cannot rule out South Korea as the geographic origin of North American EAB since the common mtDNA haplotype that is shared by all Chinese and North American EAB individuals exists in each of the Korean populations sampled. Thus, it will be necessary to increase our sampling of Asian populations if we hope to make valid inferences about the geographic location(s) of source populations that gave rise to the North American EAB infestations. To this end, collections will be conducted over the summer of 2006 in South Korea, China, and Japan to expand our EAB sample from its native range. We are also working to obtain DNA sequences from the nuclear genes wingless (Wg), phosphoenolpyruvate carboxykinase (PepCK), cytochrome c (*Cytc*), and elongation factor- 1α (EF- 1α). Finally, microsatellite markers are being developed for EAB to incorporate these highly polymorphic markers into the data set. We expect that analysis of the expanded data set will improve resolution of the genetic structure of EAB populations and allow us to determine which populations are most closely related to each other and if there was a single introduction or multiple introductions of this pest.