Distinguishing between wild capture and SIT produced Mediterranean fruit fly, *Ceratitis capitata*, using the *Hae*III Restriction Endonuclease

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The Mediterranean fruit fly, Ceratitis capitata, is native to Sub-Saharan Africa and is considered one of the most destructive agricultural pests worldwide. Methods for reducing infestation levels and minimizing medflies from establishing in a new region includes use of the Sterile Insect Technique (SIT), the release of a large number of sterile male flies into an area where medfly is detected. Lab reared flies are marked with a dye prior to releasing to distinguish SIT flies from wild flies recovered from traps. The dye occasionally fades or is rubbed off over time. It is necessary to be able easily differentiate between lab reared and wild flies in order to assess infestations. We examine the utility of Restriction Fragment Length Polymorphisms (RFLP) for differentiating between wild and Vienna 8 (-invD53)/Toliman99 lab reared flies. We examine sequences in-silico for the presence or absence of HaellI restriction site. A total of 282 medflies from eighty two different countries were examined with 40% (113 wild flies) being gathered from Central America. All wild flies were sequenced. The HaeIII restriction site was observed in 9 (3.2%) of wild flies, with two originating from Central America, four in Spain, and three from Madiera. Flies from five SIT facilities were then examined for the presence of the HaeIII marker. The restriction site was observed 100% of the time in four of the facilities. The marker was absent 40% of the time in the facility located in Hawaii. However, even with that discrepancy, the Haelll restriction site has shown some utility for distinguishing Vienna-8 from non-Vienna-8 flies suggesting it may have value for screening colonies as part of quality control and identification of captures suspected to be SIT flies.