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SEX DETERMINATION IN THE MEDFLY: A MOLECULAR APPROACH

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The Mediterranean fruit fly, *Ceratitis capitata* (Medfly), has become an important agricultural pest in many countries which economically depend on the production of fruits. The control of agricultural pests, such as the Medfly, by insecticides has unfortunately serious ramifications for the natural environment and the human health, more so than was expected previously. Hence new strategies of control are needed if one wishes to avoid the use of pesticides and their potential dangers to the environment. An attractive alternative would be a modification of the well known SIT (sterile insect technique) whereby only males would be released. In fact both the damage caused by the released sterile females with their ovipositor and the preferential mating between the released flies instead of between the released and the wild type should be overcome by a modification of the sterile insect technique, whereby only male flies would be released. Our aim is the development of a *Ceratitis* sexing strain by introducing a conditional female-specific lethal factor into the genome of the Medfly, so that in appropriate conditions there will be produced only males to sterilise and subsequently release. This modified version of the SIT will significantly improve already existing eradication programs as the costs of rearing and the disadvantages resulting from female release would be greatly reduced. Essential to this objective is a detailed knowledge of the mechanisms that control sexual development in the Medfly. One approach could be based on the knowledge available from another fruit fly *Drosophila melanogaster,* a model organism for genetics and molecular studies. By this way, it has been possible to isolate in *Ceratitis* various *Drosophila* homologous genes, as well as to develop a gene transfer technique similar to the *Drosophila P*-element method. For this purpose we searched for sex-specifically regulated genes in medfly, homologous to those involved in the *Drosophila* sex determination. Their regulative sequences should be useful in driving female-specific expression of a chimeric transgene coding for a conditional lethal function, such as the ice nucleation gene *inaZ* of *Pseudomonas syringae.* A cold shock exposure should lead to lethality by freezing those cells expressing the nucleation protein, whose activity is to order the water molecules facilitating ice formation. Because of the availability in *Drosophila* of sex-specifically regulated genes already cloned, such as *transformer* that undergoes sex-specific alternative splicing, we are using *Drosophila* transgenic flies as a preliminary step to test the functionality of this system. We have made a genetic transformation construct containing the *Drosophila transformer* gene in fusion with the *inaZ* gene so that a female-specific splicing of the pre-mRNA will lead to transcripts coding for a TRA-INAZ protein only in the female flies. Germ-line injections of this construct led us to obtain transgenic flies and to study molecularly the post-transcriptional regulation of the transgene. In the aim of identifying in *Ceratitis* gene regulatory sequences able to "drive" sex-specific expression of a transgene, we have isolated a *Drosophila doublesex* homologue gene *(Ccdsx)* producing different transcripts in the two sexes by alternative splicing and, using molecular subtractive techniques, two *Ceratitis* genes that are transcriptionally expressed at much higher levels in one sex respect to the other.