

GENOME SEQUENCING AND MUTAGENESIS IN THE PLANKTONIC DIATOM *PSEUDO-NITZSCHIA MULTISTRIATA*

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Pseudo-nitzschia multistriata is a planktonic diatom that regularly blooms in the Gulf of Naples (Mediterranean Sea). It is a toxin-producing species, as a number of strains have been shown to produce domoic acid, although it has never been linked to harmful events. *P. multistriata* has a typical size reduction-restitution life cycle and a heterothallic mating system. The ecology, life cycle and population genetics of this species have been studied intensively in the past years. Because of its ecological relevance and because of its life cycle characteristics, we propose to establish *P. multistriata* as a genetic model that could be used to study gene function in a planktonic organism through classical genetic approaches. To show that the genetics of this species can be controlled, we initiated a breeding program and produced a pedigree with four generations. We defined the minimal generation time, verified Mendelian inheritance of microsatellites markers and started a project to sequence the genome of *P. multistriata*. Initial results produced using the Illumina/Solexa platform, indicate that the genome is AT-rich and has a size of about 100 Mb. Further rounds of sequencing are ongoing to refine the assembly.

In addition, we are generating transcriptomes from strains of opposite mating types above and below the sexual size threshold and from sexual stages. These experiments, designed to explore the molecular mechanisms regulating the life cycle and sexual reproduction in this species, will reveal the genetic bases of the mating system and at the same time will aid in the annotation of the *P. multistriata* genome.

Finally, we are using chemical and UV mutagenesis to generate stable mutants and are designing a forward genetic screen which will involve identification of mutants for domoic acid or fatty acid production. Availability of the genome sequence will be exploited to identify the genes responsible for phenotypes of interest, either by positional cloning or by genome resequencing.

Through this project, we aim to consolidate *P. multistriata* as a model organism for molecular, genetic and biotechnological studies, complementing existing diatom models.