

# Stable Fluorescent Mutant of Perkinsus marinus Expressing GFP

### **Summary**

Perkinsus are intracellular parasites that cause disease in shellfish, especially bivalve molluscs. Its infection has devastated oyster

## **Key Investigator**

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#### Field Research tool

Tangible research property

**Technology** Mutant protozoan

## Advantages

GFP-tagged *P. marinus* allows easier visualization and quantification of infection in bivalves

GFP-expression is induced early and stably, lasting longer than 8 months

## Status

Available for non-exclusive licensing

#### UMB Docket Reference GV-2014-015

## **External Reference**

Fernández-Robledo JA et al (2008) *Molec and Biochem Parasitology* 157:44-53. populations along the US coasts. In addition to the economic value of shellfisheries, filterfeeding bivalves such as oysters play a critical role in maintaining water quality, making *Perkinsus* a serious threat to the economy and the health and integrity of the coastal ecosystem. Numerous attempts to control Dermo disease, a result of *P. marinus* infection, have been unsuccessful to date, partly due to a lack of information about fundamental aspects of *P. marinus* biology. UMB researchers have developed a transfection system for Perkinsozoa and created a stable, fluorescent mutant of *P. marinus* that expresses GFP. This transgenic organism can be used to study the mechanisms underlying *P. marinus* infection in bivalves, allowing for easier visualization and quantification.

#### Market

Bivalves such as oysters are important both ecologically and economically. As the causing agent of Dermo disease in oysters, *P. marinus* has caused mass mortality of Eastern oysters commonly cultured and wild-harvested as food species. Found along the eastern coast of the USA, from Maine to Florida, the presence of this parasite has severely reduced the abundance and productivity of oysters, particularly that of *Crassostrea virginica*. In the Mid-Atlantic region, *P. marinus* is responsible for a decrease of at least two-thirds of the surplus production available to fisheries. GFP-labeled *P. marinus* can help better understand the biology behind its infection and assist in identifying successful intervention strategies to curb it, both crucial to protecting the ecosystem and aquaculture industries.

### Technology

Using optimized *P. marinus* culture methods, UMB researchers constructed a vector based on a highly expressed *P. marinus* gene tagged with GFP (pPmMOE-GFP). Under optimized transfection conditions, exogenous pPmMOE-GFP DNA was introduced into *P. marinus* trophozoites by electroporation with a robust efficiency of transfection of ~38%. Fluorescence can be detected starting 14 hours after electroporation and stably expressed for longer than 8 months. The visualization allows researchers to study its location, growth rate, and proliferation.

## **Technology Status**

pPmMOE-GFP-tagged *P. marinus* has been tested *in vivo*, where PmMOE-GFP is expressed with a strong concentration in the inner lining of the plasma membrane.