

Targeted-metabolomics of phytoplankton blooms in the Gulf of Naples and effects of algal oxylipins on the reproduction and gene expression of copepods

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Project Summary/Abstract

Several diatom species produce cytotoxic oxylipins that impair reproductive and developmental processes in their main grazers, crustacean copepods, and ultimately reduce their recruitment at sea. Traditionally, the diatom *Skeletonema marinoi* and the copepod *Calanus helgolandicus*, both occurring in the Northern Adriatic Sea, have been used as model system in studies on chemically-mediated diatom-copepod interaction. Comparatively fewer studies have been performed on other co-occurring diatom and copepod species in the Mediterranean Sea. Reduced hatching success, for example, has been reported in the copepod *Temora stylifera* from the Gulf of Naples, in presence of the oxylipin-producing diatoms *Thalassiosira rotula*, *Skeletonema pseudocostatum*, *Chaetoceros affinis* and *Chaetoceros socialis*, which are recurrently identified in phytoplankton blooms of the Gulf of Naples. Recently, new cytotoxic oxylipins have also been identified in species of the genus *Leptocylindrus* from the same area, but these diatoms have never been tested on copepods. Despite the occurrence of several oxylipin producing diatoms in the Gulf of Naples, and their possible impact on co-occurring copepod species, however, a detailed analysis of oxylipin production during phytoplankton blooms in this area has never been attempted. The phytoplankton seasonal dynamic in the Gulf Naples is characterized by well-diversified phytoplankton communities, with high fluctuations in terms of species composition and abundances over time. This complexity pose a major challenge to the understanding of the impact of algal metabolites in prey-predator interactions. A comprehensive inventory of phytoplankton signals and large scale approaches addressing the system as a whole, might, thus, lead to elucidate the ecological role of algal infochemicals on plankton dynamic in this area. The general goal of this PhD project is to use a targeted-metabolomic approach to investigate metabolic changes in the whole phytoplankton community of the Gulf of Naples, and its possible impact on the reproductive and molecular responses of co-occurring selected copepod species from the same area. Special attention will be paid to lipid profiling with a major focus on diatom oxylipins.

Specific objectives are:

1. To investigate the metabolic profile of defined groups of chemically characterized metabolites, such as oxylipins, produced during phytoplankton blooms in the Gulf of Naples. Phytoplankton samples will be collected at the fixed coastal station Long Term Ecological Research Station MareChiara (LTER-MC) and analyzed by hyphenated analytical methods (e.g. LC-MS/MS and GC-MS/MS) of crude extracts or fractions.
2. To investigate the *in situ* reproductive and molecular responses of copepod species sampled at LTER-MC, during a one year seasonal cycle. The species selected are *T. stylifera* and *C. helgolandicus*, which occur during autumn-winter and spring, respectively. Reproductive and molecular responses of the copepods will be correlated with both metabolomic data (Ob. 1), and environmental parameters measured at LTER-MC.
3. To investigate the reproductive and molecular responses of *T. stylifera* and *C. helgolandicus* females fed on selected oxylipin-producing diatoms from the Gulf of Naples. The diatom species will be selected according to the literature and depending on the results observed at sea (Ob. 1 and Ob. 2).

The intellectual merit of the proposed activity is to provide, for the first time, a field study of biologically annotated metabolites, e.g. oxylipins, during plankton blooms and a comprehensive analysis of the reproductive and molecular responses of important copepod species both *in situ* and in the laboratory. Our PhD proposal will add new knowledge on the occurrence of chemically-mediated prey-predator interactions in the Gulf of Naples, and possibly, on their role in shaping the structure and functions of this strategic coastal area of the Mediterranean Sea.

The broader impacts resulting from the proposed activity are in the context of 1) biodiversity and ecosystem functioning: to provide a chemical fingerprinting of phytoplankton blooms and correlate major metabolic pathways with bloom successions and consumer fitness; to contribute in bridging the gap between mechanistic molecular understanding and prey-predator responses; 2) ecosystem health and sustainable aquaculture: to promote the development of molecular markers in copepods, to be used in environmental risk assessments in relation to harmful algae; to stimulate the use of algal infochemicals in 'push-and-pull' strategies in integrated pest management in aquaculture. Increasing our understanding on the chemical ecology of food-web networks would provide opportunities for implementing new management solutions, improving the predictive capability of environmental change scenarios and sustaining healthy marine resources.