Phytoplankton Community Dynamic: A Driver for Ciliate Trophic Strategies

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INTRODUCTION

"The understanding of the temporal dynamic of primary grazers channeling energy and carbon from primary producers is important for evaluating aquatic ecosystems functioning... This study aims to assess the temporal coupling between phytoplankton and its protist grazers in a temperate mesohaline estuary (Roskilde Fjord, Denmark), evaluating differences in potential grazing rates of distinct trophic strategies over different time scales. More specifically, this study seeks to answer the following questions: (1) Are ciliates the dominant pelagic grazers in Roskilde Fjord (RF)? (2) Do mixotrophic ciliates comprise a significant proportion of the total ciliate biomass and thereby contribute significantly to the transfer of energy to higher trophic levels?"

FlowCam METHOD

"Phytoplankton was analyzed with a pulse shape recording flow cytometer, whereas ciliates were

analyzed by a color FlowCam... Ciliate abundances and body volumes were analyzed from live samples using a color FlowCam... FlowCam allowed for identification of different ciliate morphotypes, and for the identification of prey items inside many of the ciliates"

RESULTS & CONCLUSIONS

"The use of in-flow techniques supports the analysis of phytoplankton and their microzooplankton grazers with high frequency...Ciliates are likely the main pelagic grazers in RF and probably play an essential role in the food web, linking primary production of small-celled organisms to higher trophic levels."

Dynamics of transparent exopolymer particle production and aggregation during viral infection of the coccolithophore, *Emiliania huxleyi*

Environmental Microbiology (2018) doi: 10.1111/1462-2920.14261

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INTRODUCTION

"Emiliania huxleyi's extensive oceanic blooms are often terminated by coccolithoviruses (EhVs) with the transport of cellular debris and associated particulate organic carbon (POC) to depth being facilitated by transparent exopolymer particles (TEP)-bound 'marine snow' aggregates. The dynamics of TEP production and particle aggregation in response to EhV infection are poorly understood... Using flow cytometry, spectrophotometry, and FlowCam visualization of alcian blue (AB)-stained aggregates, we assessed TEP production and the size spectrum of aggregates for E. huxleyi possessing different degrees of calcification and cellular CaCO₃:POC mass ratios, when challenged with two EhVs."

FlowCam METHOD

"Recent advances in FlowCam technology allows direct visualization and characterization (size spectrum, shape, association with cells) of AB-stained TEP particles... FlowCam imaging directly characterized the number, size and blue:red ratio of Alcian Blue (AB)-stained, polysaccharide containing aggregates for infected and uninfected *E. huxleyi* cells."

RESULTS & CONCLUSIONS

"Our work shows the potential for EhV infection to trigger aggregation and serve as mechanistic context to help explain observed TEP production, aggregation, and high PIC and POC fluxes of EhV-infected blooms in the North Atlantic."



FlowCam optimization: Attaining good quality images for higher taxonomic classification resolution of natural phytoplankton samples

Limnol. Oceanogr.: Methods (2016) doi: 10.1002/lom3.10090

M. Camoying, A. T. Yñiguez

INTRODUCTION

"This study aimed to produce high resolution FlowCam images of fixed phytoplankton samples from natural environments without compromising sample analysis time. It was also aimed to optimize the capability of FlowCam's VisualSpreadsheet software to automatically classify phytoplankton images."

FlowCam METHOD

"Samples were analyzed in Autoimage Mode... To evaluate the counting accuracy of the determined optimum software and hardware settings (flow cell and objective combination), FlowCam phytoplankton counts were compared to that of the microscopy method.... To optimize the auto-classification functionality of FlowCam, "statistical filters" were built for each phytoplankton library and were assessed for accuracy in filtering images of the target group."

RESULTS & CONCLUSIONS

"The use of FOV300 flow cell and 10X objective

combination has proven to be effective in producing good quality images at a faster rate. The modified hardware configuration resulted to FlowCam counts that were comparable to that of the standard microscopy method. FlowCam was able to automatically classify images of dominant phytoplankton groups in the two key sardine fishery areas in the Philippines with relatively high accuracy values. These phytoplankton groups are represented by genera with complex morphological structures (e.g., setae) such as Chaetoceros and Bacteriastrum as well as those genera with simple shapes such as Pseudo-nitzschia (thin-elongate) and Coscinodiscus (spherical)."



Fig. 2: Relationship between phytoplankton counts from FlowCAM and microscopy (Camoying & Yñiguez)

Note from Fluid Imaging Technologies – The authors recommend a configuration of the 10X objective with the 300 μ m 'Field-of-View' (FOV) flow cell. While such a configuration can be used with the FlowCam, we recommend that the 10X objective be coupled with the 100 μ m FOV. While a larger flow cell does provide for a quicker sample processing time, the quality of particle images may be compromised. The author's used a VS-4 model in their study. Today we offer a new FlowCam model (FlowCam 8000) that has a camera with a field of view three times larger than the VS-4. The larger field of view provides for a much quicker sample processing time.



A new method for acquiring images of meiobenthic images using the FlowCam

MethodsX (2018) doi: 10.1016/j.mex.2018.10.012

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INTRODUCTION

"The purpose of this study was to develop a new for investigating sediment-inhabiting method meiobenthos using the Flow Cytometer And Microscope (FlowCam)... Meiobenthos cannot be examined in the same manner as plankton using the FlowCam system due to sediment contamination... Sediment samples typically contain more sediment particles than meiobenthic specimens... sediment particles becoming stacked in the flow cell when... poured directly into the FlowCam as well as many more images being [acquired], which increases the volume of data and image-analysis time... Therefore, appropriate modifications are needed."

FlowCam METHOD

"Meiobenthic specimens could be separated from sediment and other debris using a centrifugal method with a high-density solution (e.g., colloidal silica at 1.15–1.18 g/cm³) to provide suitable specimens for determination by FlowCam... The separated samples were gently shaken and suspended in the colloidal silica and were then pipetted into the FlowCam system. Images of the specimens were captured using FlowCam's Auto Image mode at an Auto Image Rate of 20 frames per second. The captured images were processed as previously described using VisualSpreadsheet and were then assessed and classified to higher taxonomic levels, such as Nematoda and Copepoda... After examination with FlowCam, the samples were recollected and reexamined under a binocular stereoscopic microscope for classification and enumeration at higher taxonomic levels."



Fig. 1: Relationship between the numbers of metazoan meiobenthos observed using FlowCam and microscopy (Kitahashi et al.)

RESULTS & CONCLUSIONS "FlowCam successfully captured the images of the meiobenthic specimens in the samples, including metazoan meiobenthos and protists... and there were significant linear relationships detected between the numbers of individuals observed with FlowCam and those with microscopy."

