

YOKOGAWA 

Using the FlowCam for HAB Observations

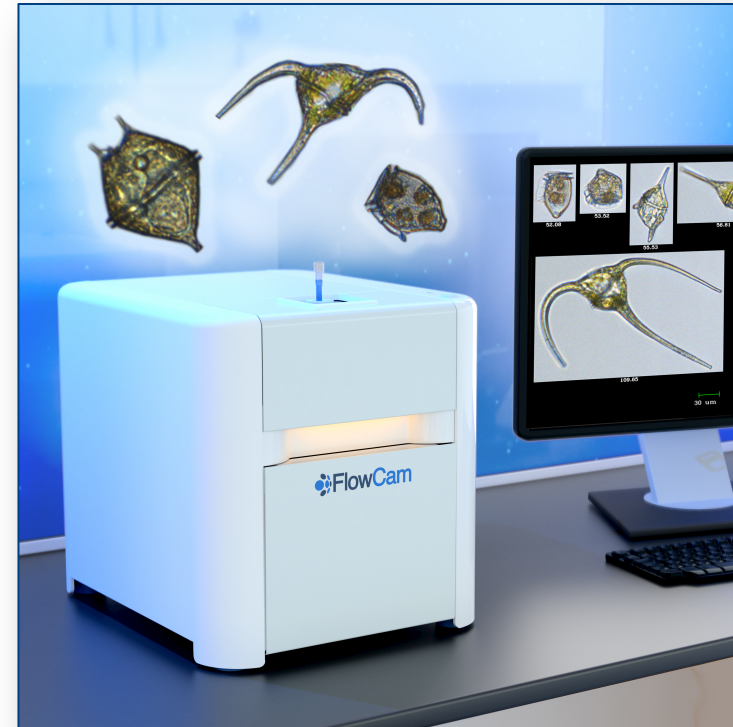
Savannah Judge & Ian Salter

GlobalHAB Symposium | August 23, 2022

 FlowCam[®]

Agenda

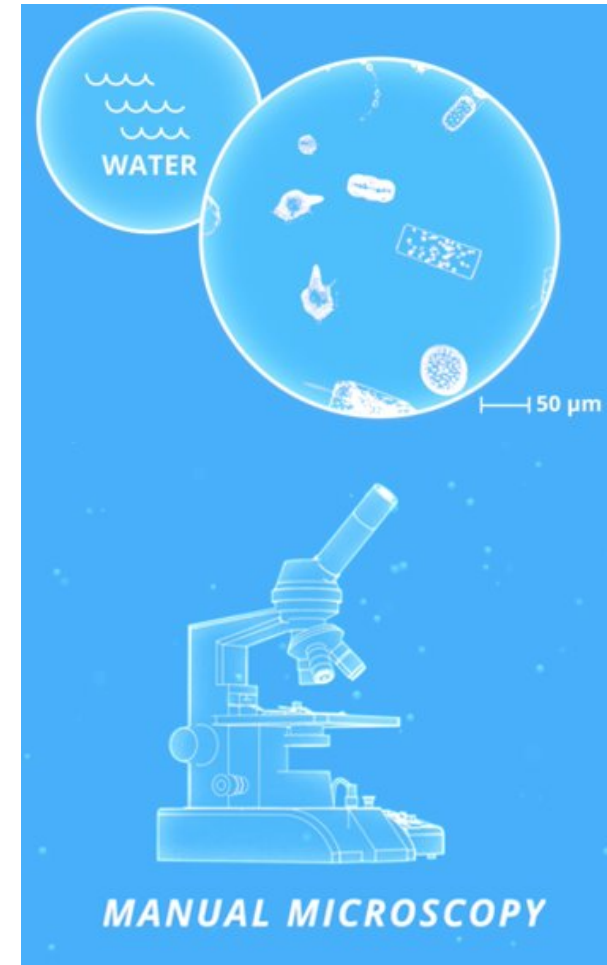
- Introductions
- FlowCam History & Overview
- Notable HAB Studies
- Case Study: Faroe Marine Research Institute
- Questions/Discussion



FlowCam 8000 with marine phytoplankton

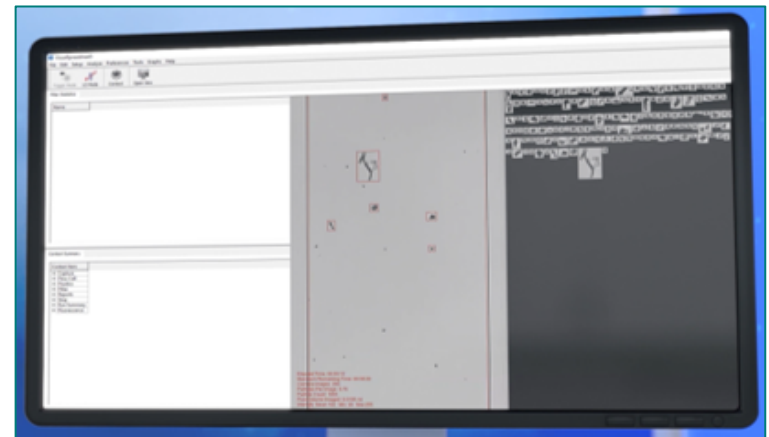
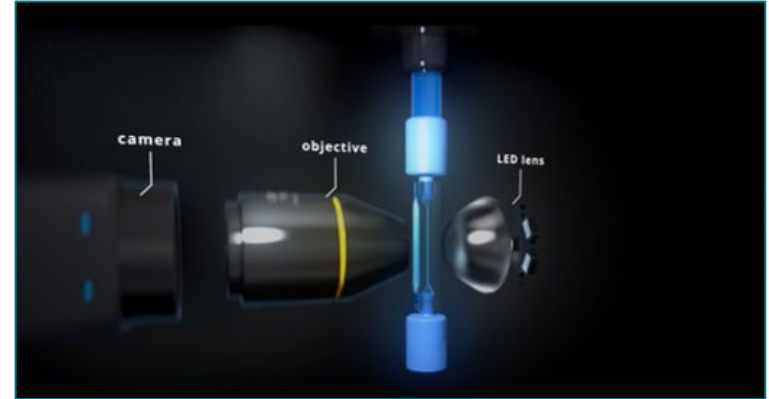
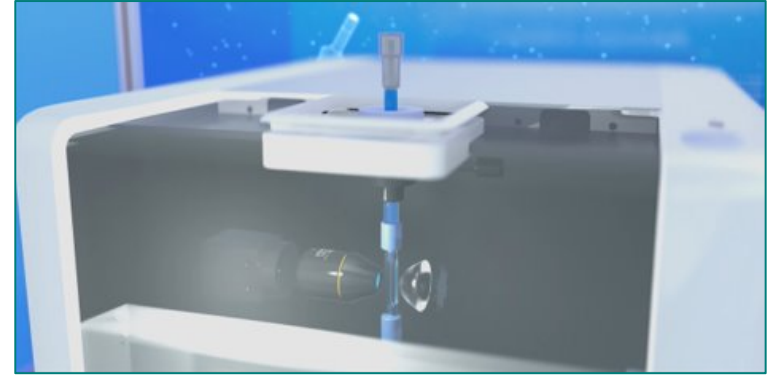
FlowCam Overview

- Flow Imaging Microscopy (FIM)
- Invented by biological oceanographers to:
 - Provide a faster alternative to manual microscopy
 - Add a visual element to flow cytometry
- Used today in a variety of fields, including aquatic research, algae cultivation, biopharmaceutical research, and more.

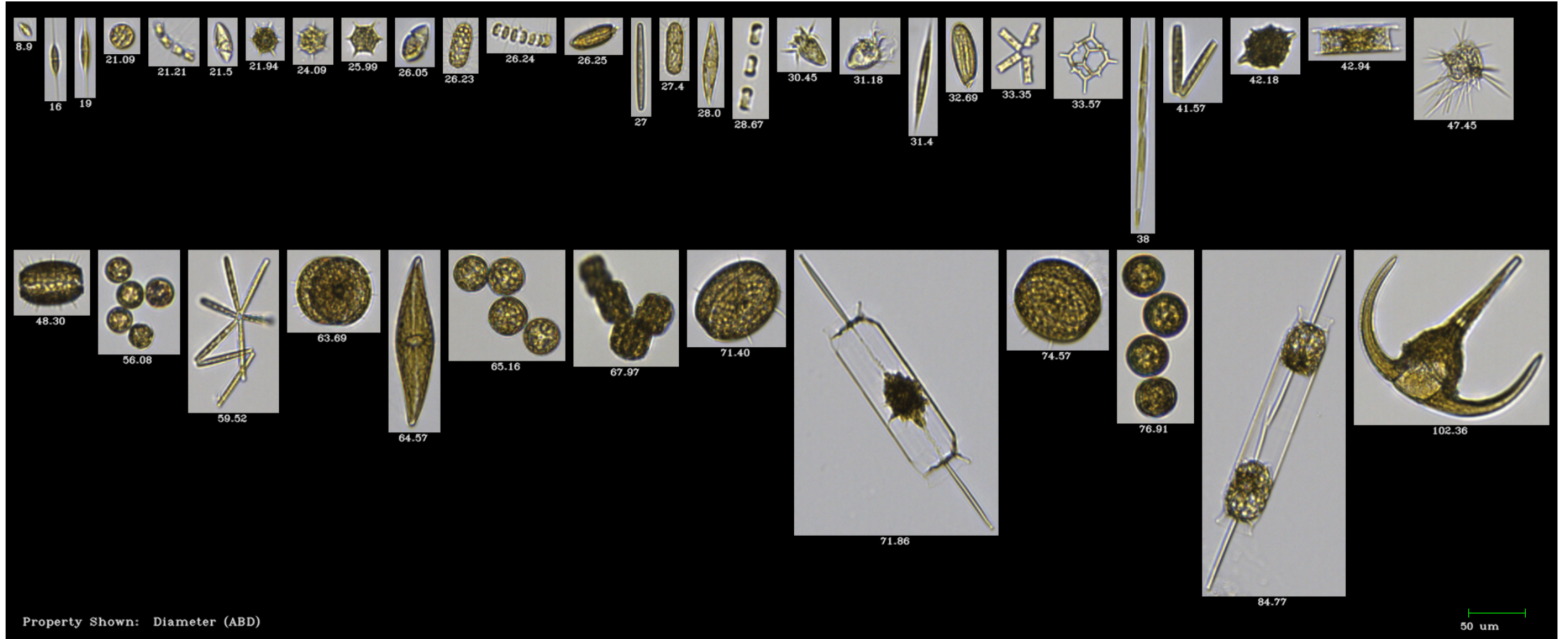


How it Works

1. Sample is filtered & introduced to the top of the FlowCam.
2. Syringe pump draws sample through the instrument.
3. Sample passes in front of an objective lens and digital camera and is backlit by an LED.
4. Software "crops" particles (ROIs), saving them to a database and recording many measurements.
5. User builds libraries, classifies particles, and exports reports.



GNATS Cruise, 2018 (10X)



FlowCam® Suite of Instruments

Particle imaging and characterization for every application



FlowCam 8000 Series

Our most versatile instrument for applications ranging from biopharma to materials to plankton analysis. Laser excitation available.
Particle size range: 2 μm - 1 mm



FlowCam LO

Flow Imaging and Light Obscuration in a single instrument. Particle size range: 2 μm - 70 μm



FlowCam Cyano

For quick cyanobacteria detection and enumeration using pigment fluorescence.
Particle size range: 2 μm - 1 mm



FlowCam Nano

Submicron particle imaging for particles 300 nm to 2 μm .



FlowCam 5000

Optimized for your application; our most affordable instrument.
Particle size range: 3 μm - 1 mm



FlowCam Macro

Analysis of visible particles from zooplankton to fibers to food and beverage ingredients.
Particle size range: 300 μm - 5 mm



ALH for FlowCam

Automated liquid handling for unsupervised analysis; compatible with FlowCam 8000, FlowCam LO, and FlowCam Cyano.



VisualSpreadsheet

Powerful software to analyze image images and visualize your results captured by FlowCam.



Biopharmaceutical Development

Flow Imaging Microscopy is a recommended orthogonal method to Light Obscuration for determining subvisible particulate content per USP <1788>. FlowCam is widely used in biotherapeutics research, formulation, and development for proteins, nano-drug delivery systems, and cell and gene therapy products.



Materials Characterization

FlowCam is employed in a wide variety of materials and chemicals-focused applications: from the characterization of food and beverage ingredients to printer toner and superabrasives, ion exchange resins, column packing material, fibers, additive manufacturing, polymer composition analysis, chemicals, cosmetics formulations, and microencapsulation processes.



Marine and Freshwater Research

For more than 20 years, scientists have been using FlowCam as an automated, fast and accurate, easy-to-use alternative to manual microscopy to monitor plankton community composition. FlowCam has become a valued instrument worldwide for studying marine and freshwater microorganisms.

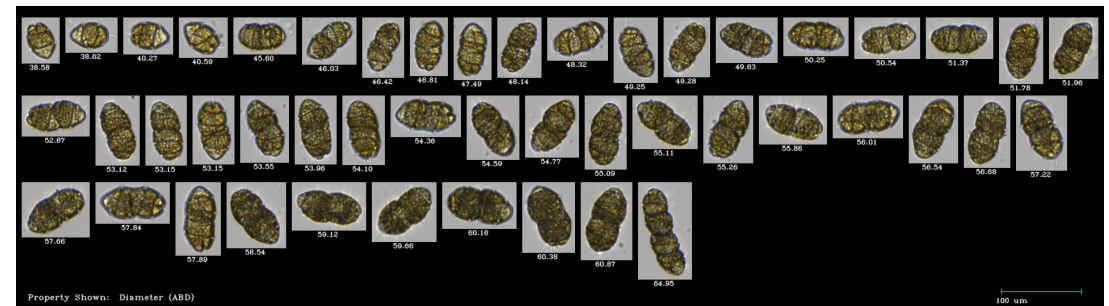
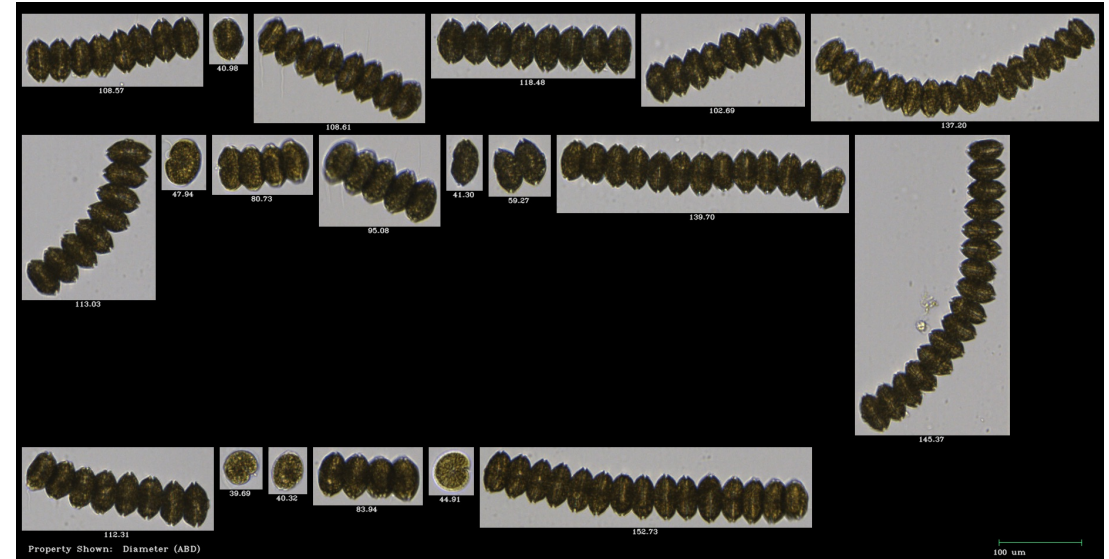


Water Quality Monitoring and Environmental

FlowCam provides a proactive, cost-effective, and scalable solution to monitor raw and treated water. Water utilities around the world use FlowCam to detect and quantify populations of taste and odor producers, filter-fouling diatoms, and potentially toxic cyanobacteria. Environmental applications include monitoring soil microorganisms, pollen, seeds, and pollutants.

Recent Examples of HAB Studies in Progress

- Virginia Institute of Marine Science: Documenting life cycle of *Alexandrium monilatum*, and tracking *Margalefidinium polykrikoides*
- Queens College: Comparing FlowCam, microscopy, and SHA
- Challenges:
 - Automating classification
 - Small & chain-forming organisms



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Flowcam observations of HABS: A case study from a Faroese fjord

Global HAB symposium on automated plankton observations
August 2022, Kristineberg



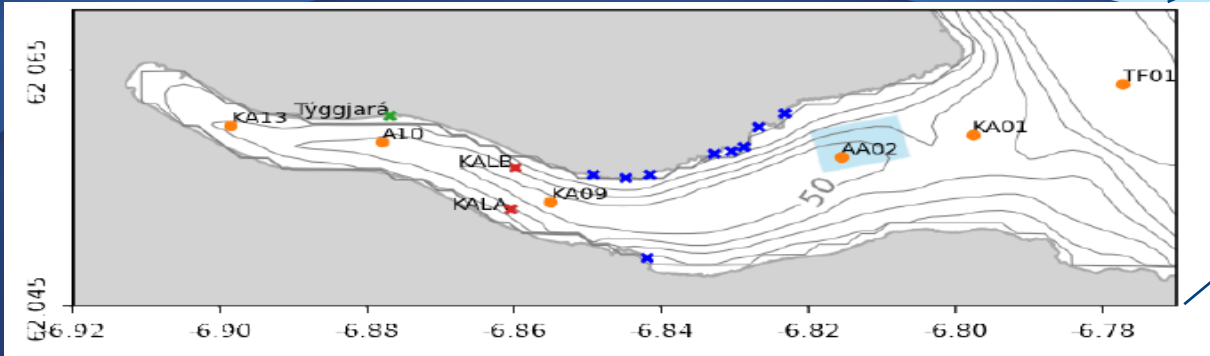
HAVSTOVAN
FAROE MARINE RESEARCH INSTITUTE

Dr. Ian Salter (Faroe Marine Institute), Asa Jacobsen (Aquaculture Research Centre)

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Flowcam observations of HABS: A case study from a Faroese fjord

Kaldback Fjord

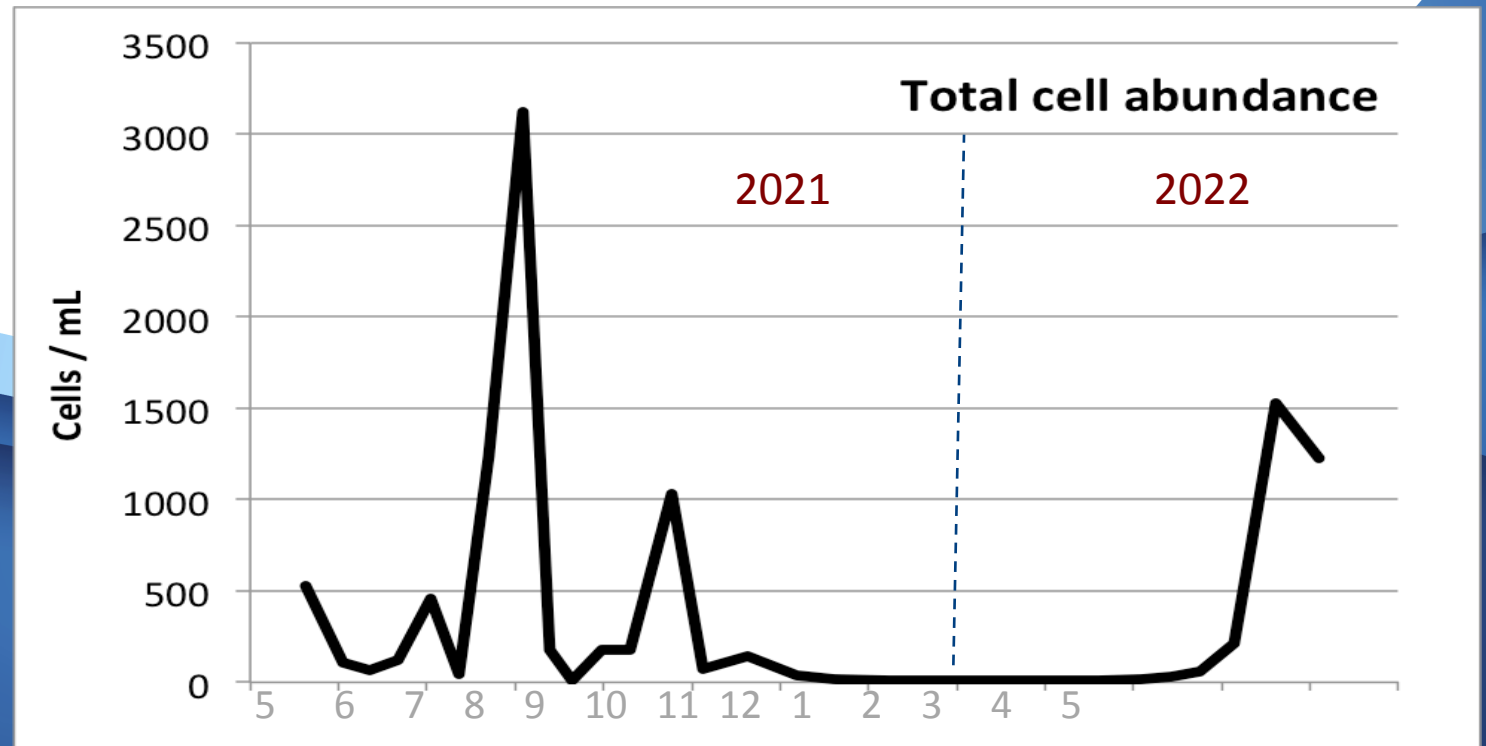


Flowcam observations of HABS: A case study from a Faroese fjord

Fiskaaling
(Aquaculture research Centre)
Asa Jacobsen

Weekly phytoplankton
Observations at A10

Detailed taxonomic counts



HAB Flowcam pilot study

Objectives

Identify potential HAB species from microscope counts.

Analyse archived samples with Flowcam 8000
4X objective, 10 mL – 0.9 mL/min (15 mins)

Compare ability of Flowcam to detect HAB species

- (i) Abundance
- (ii) Biovolume

Time/cost assessment

HAB Flowcam pilot study

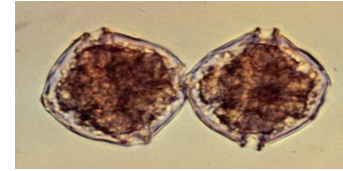
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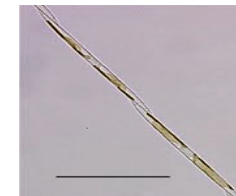
Time/cost assessment



Alexandrium spp
PSP



Dinophysis spp
DSP



Pseudo-nitzschia spp
ASP



Phaeocystis
Gill health



Chaetoceros spp
Gill health

HAB Flowcam pilot study

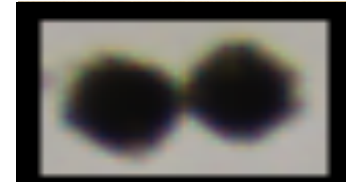
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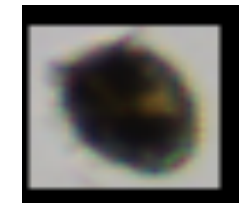
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Alexandrium spp
PSP



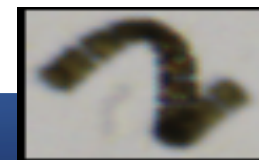
Dinophysis spp
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Pseudo-nitzschia spp
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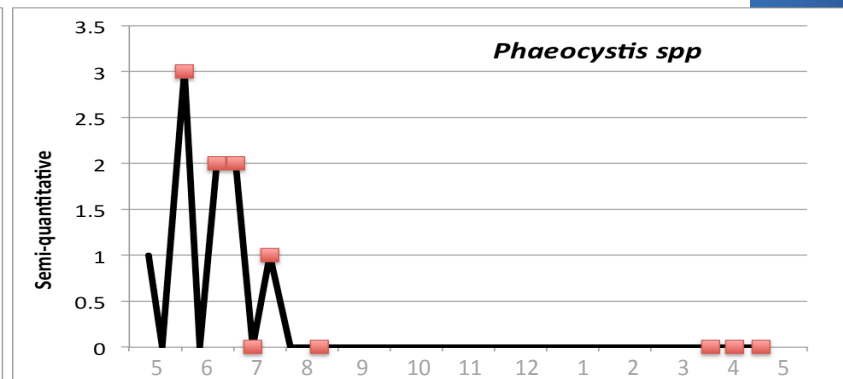
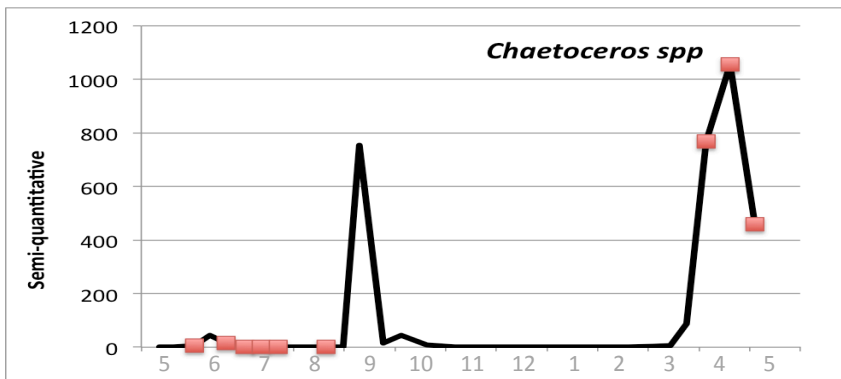
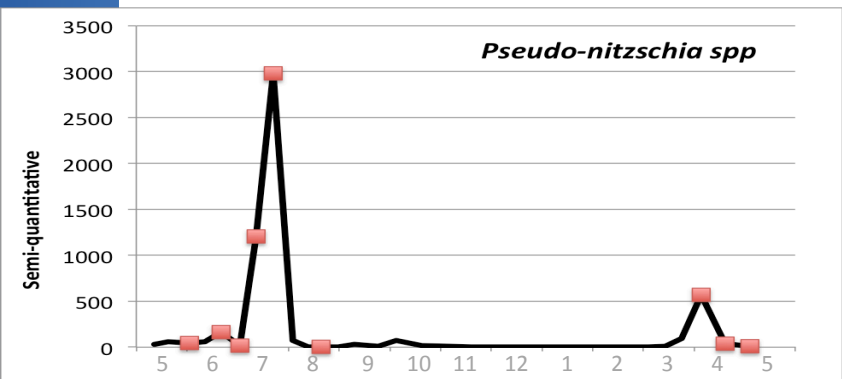
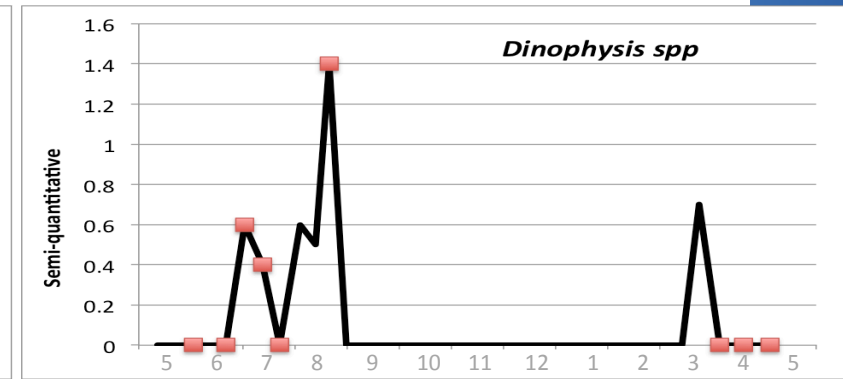
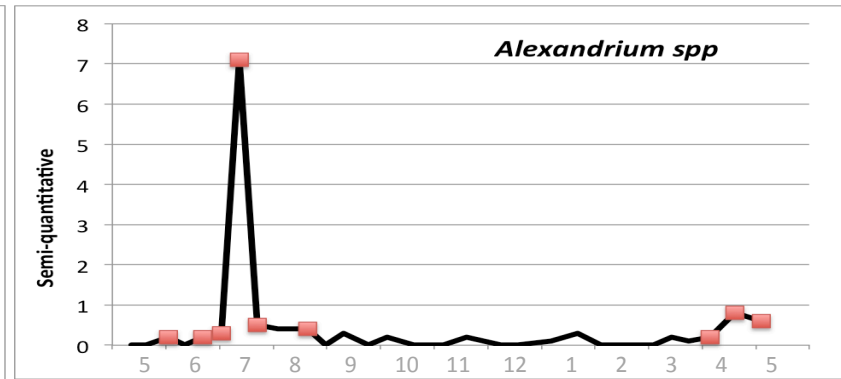
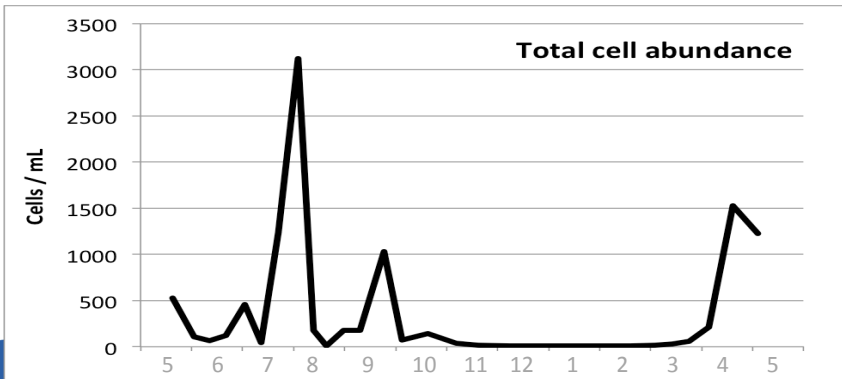


Phaeocystis
Gill health

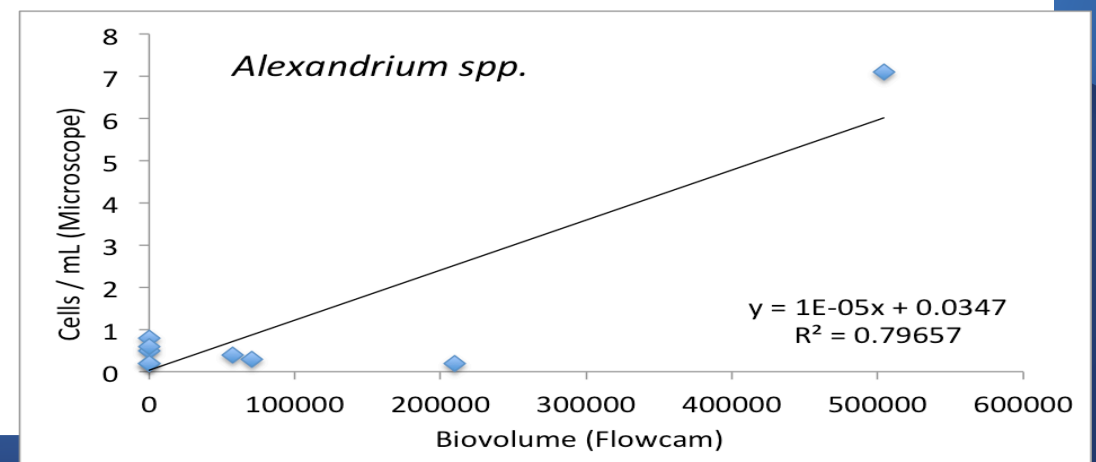
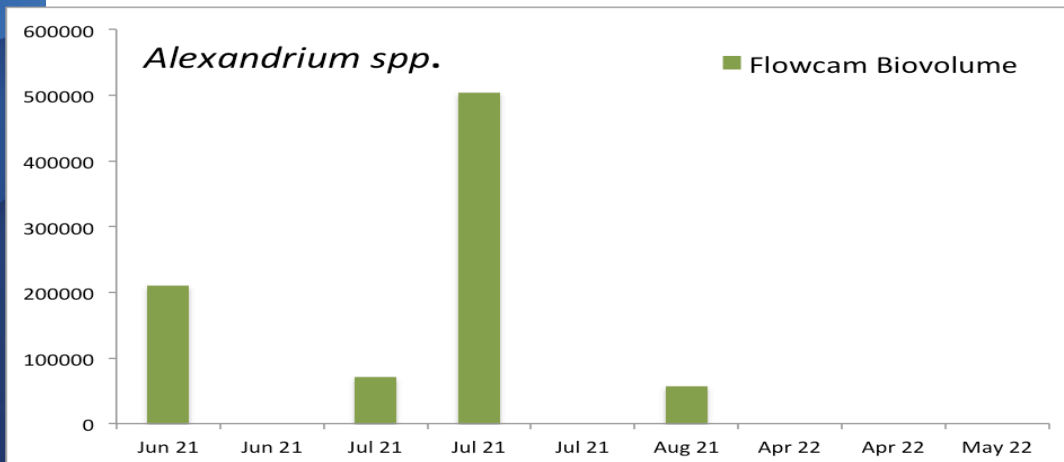
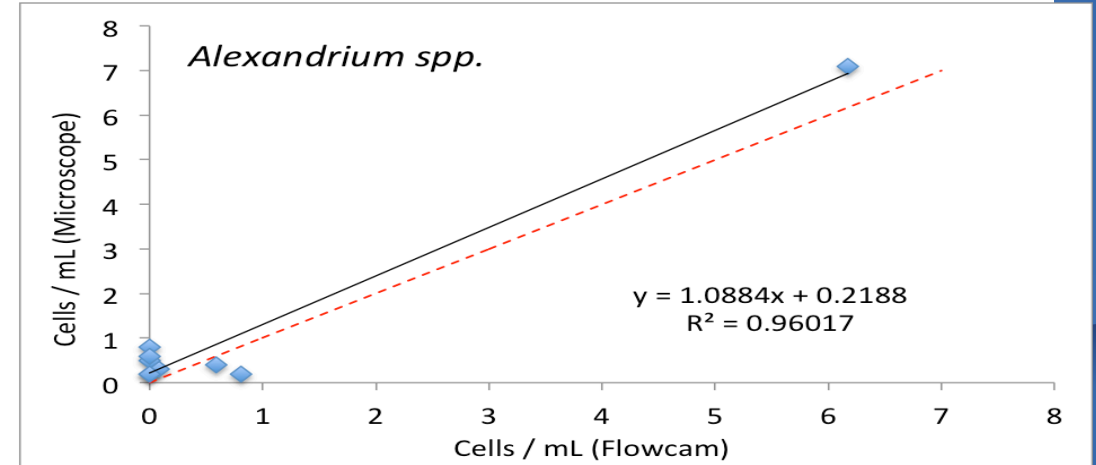
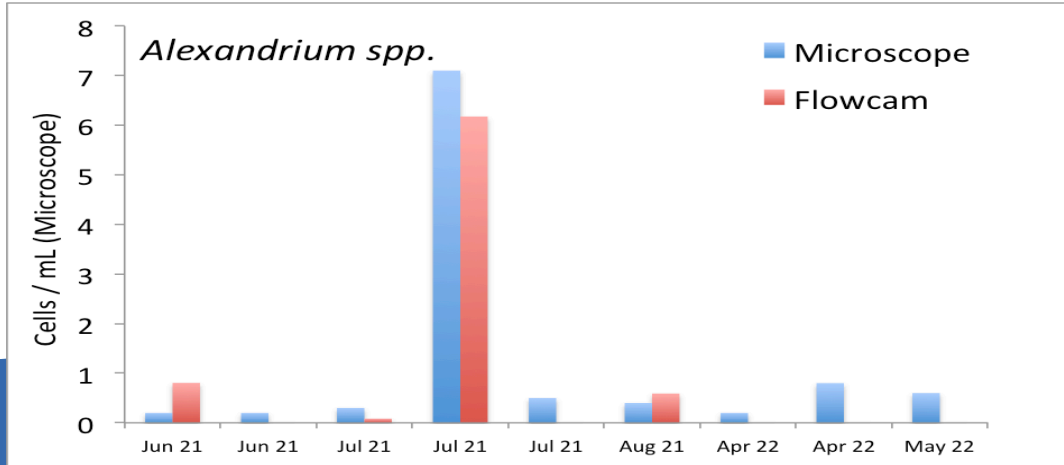


Chaetoceros spp
Gill health

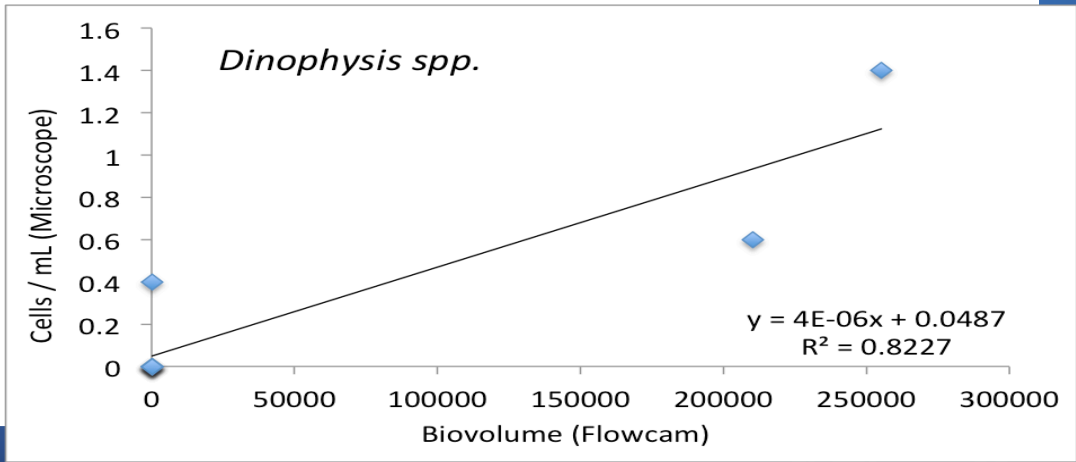
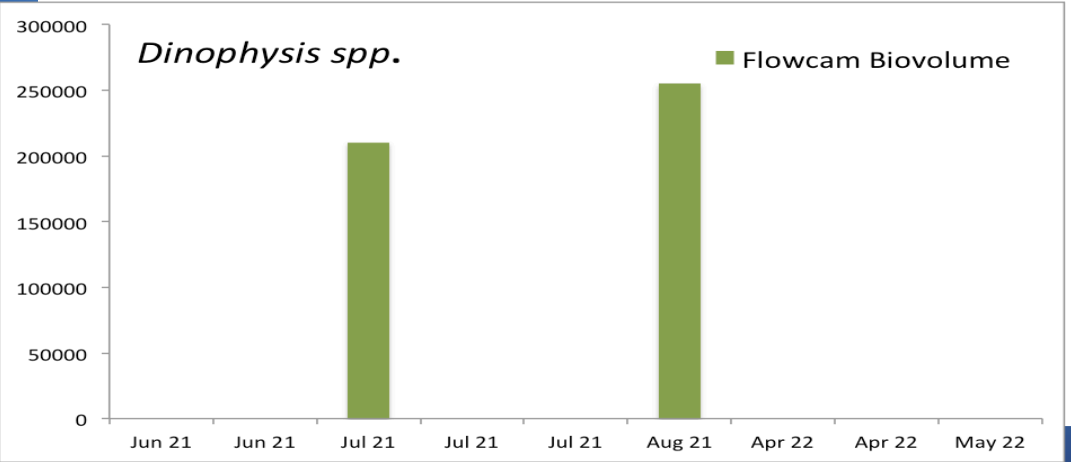
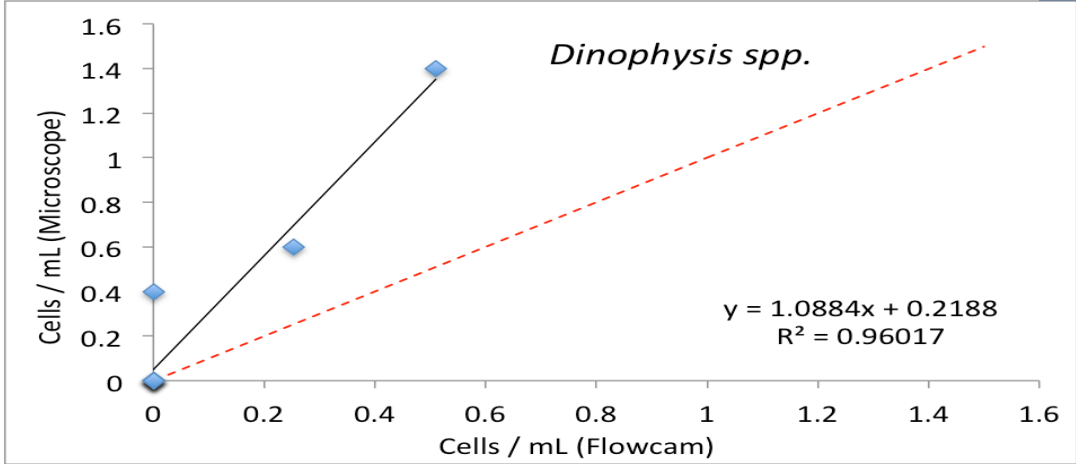
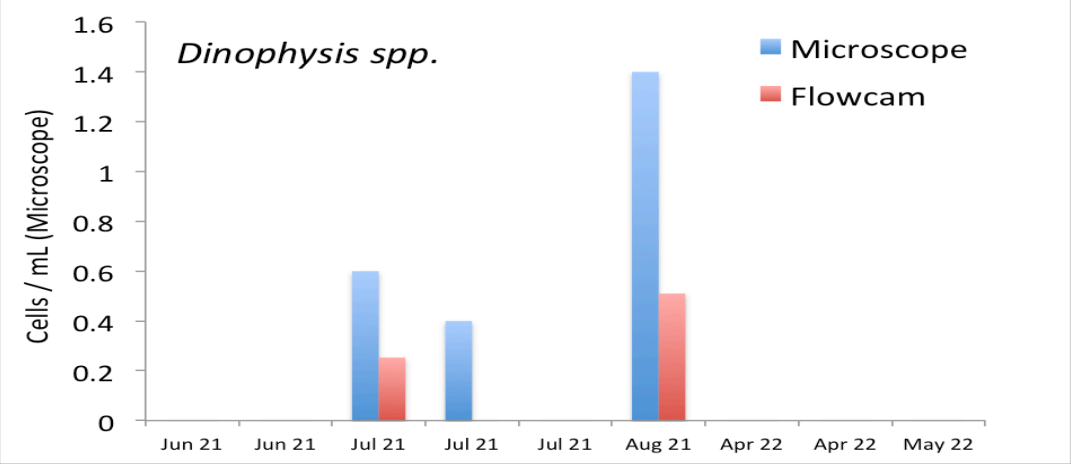
Microscope counts



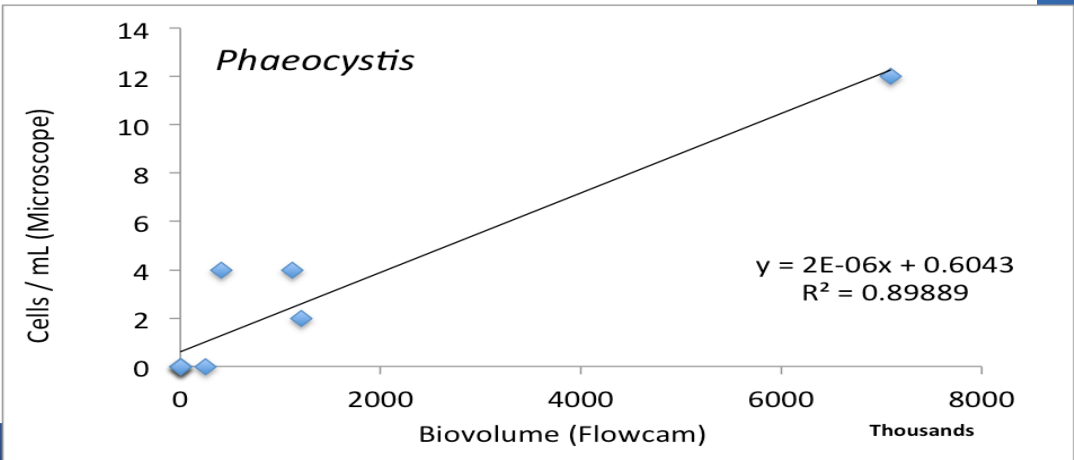
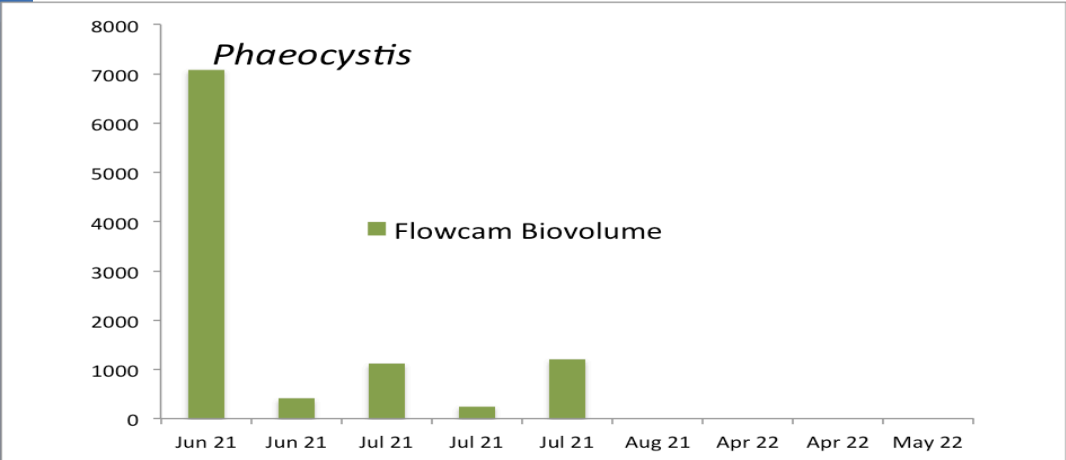
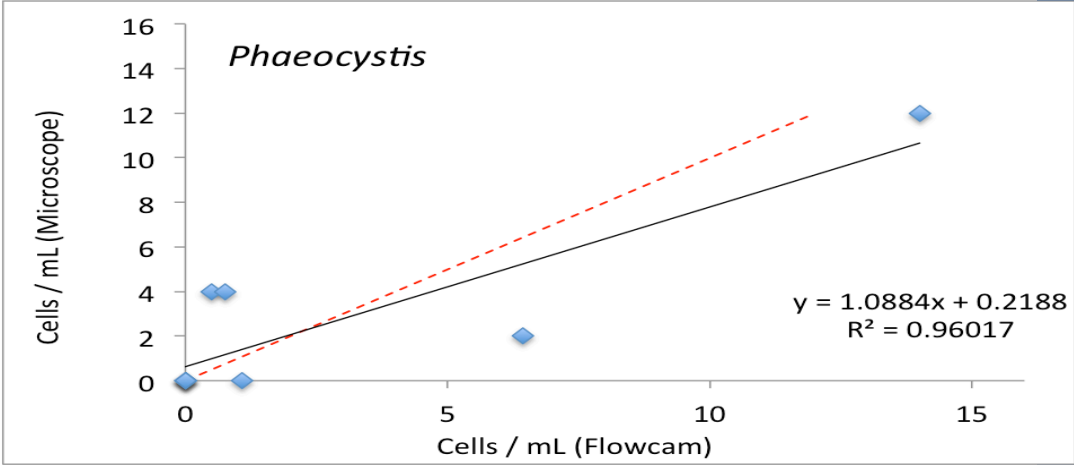
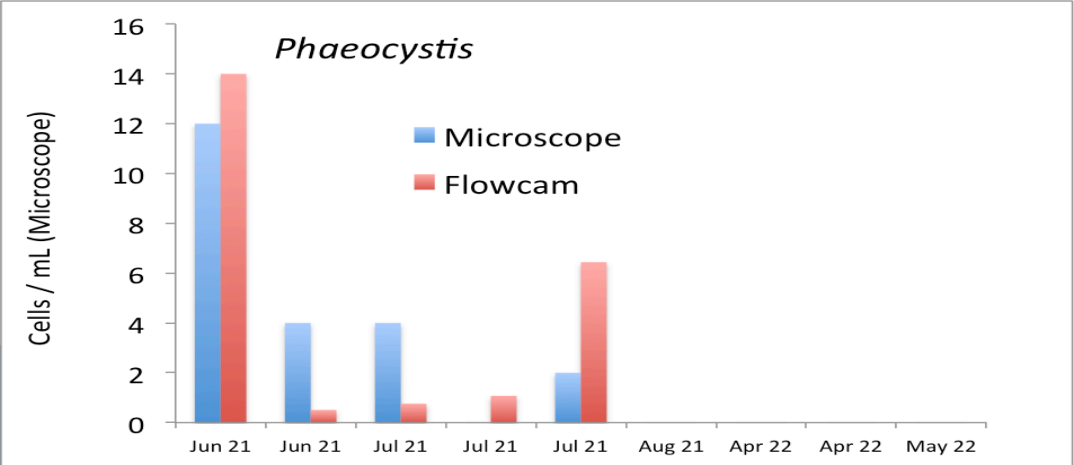
Microscope Flowcam comparisons



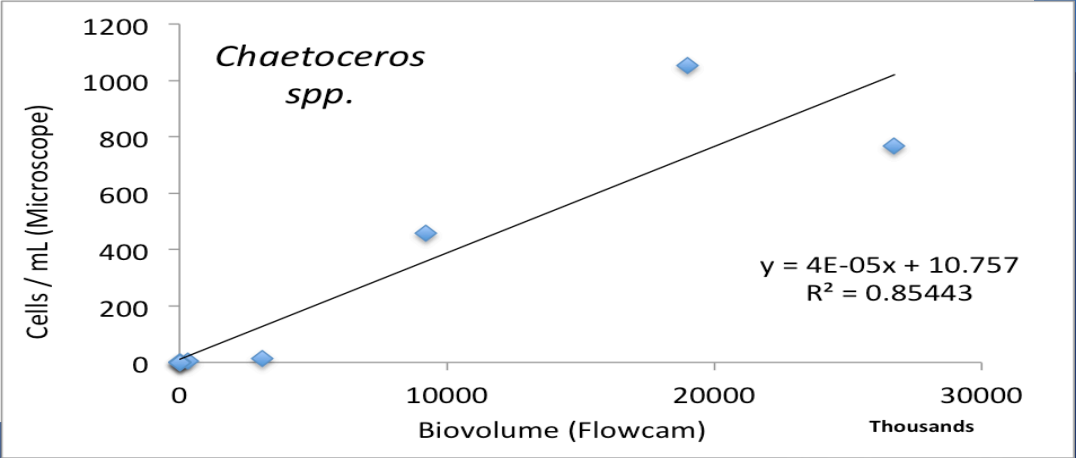
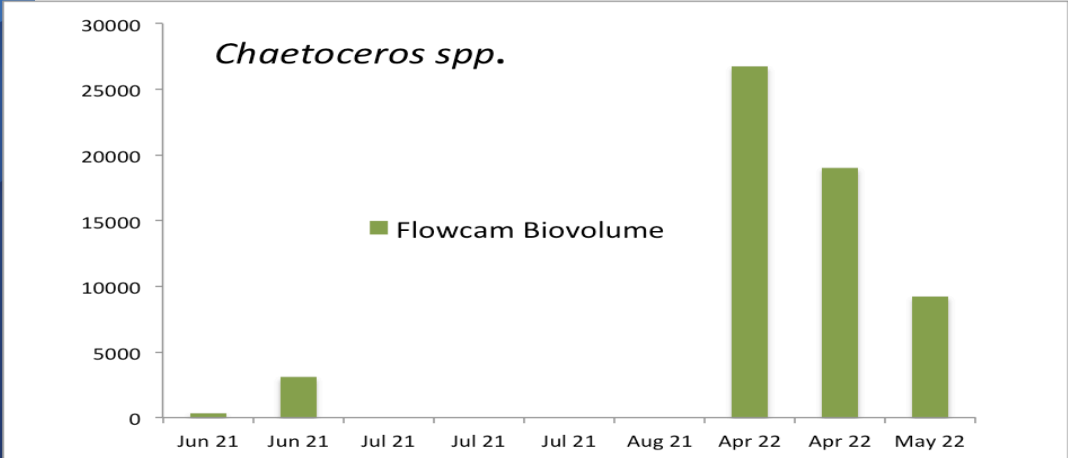
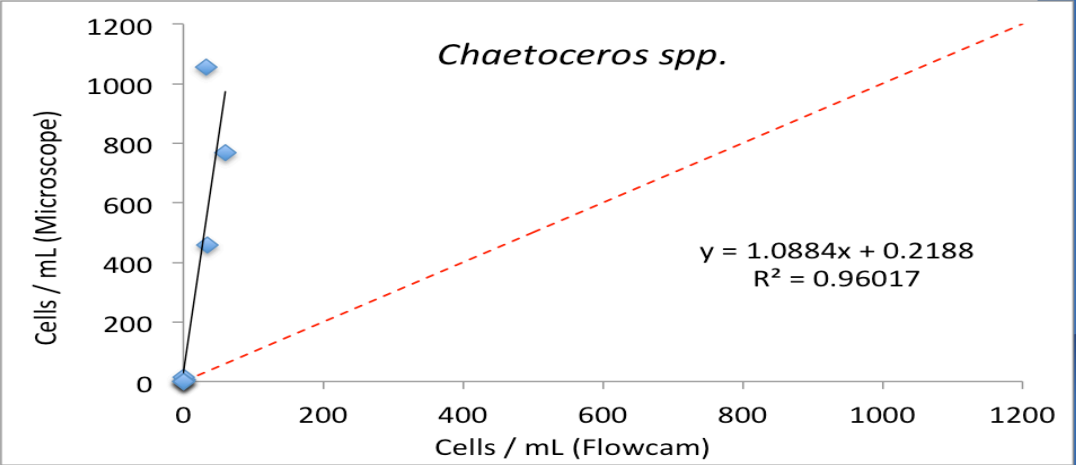
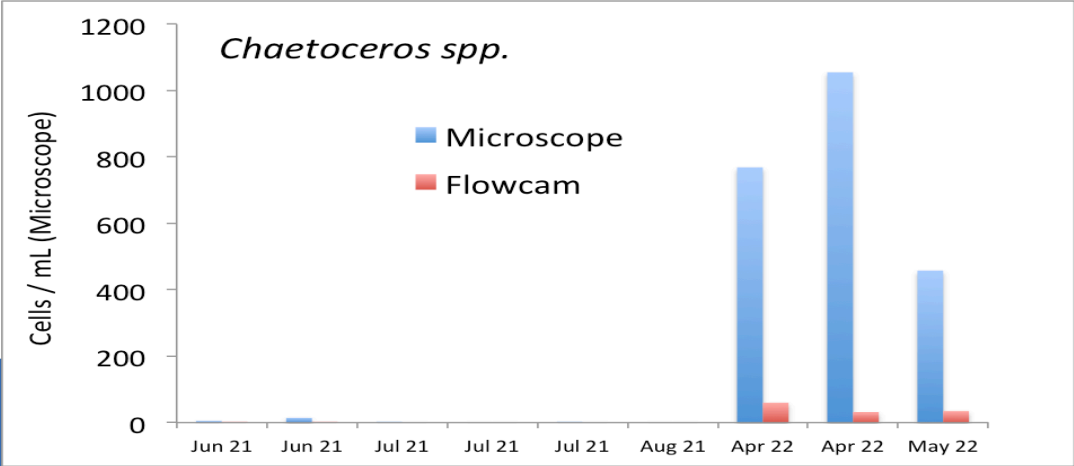
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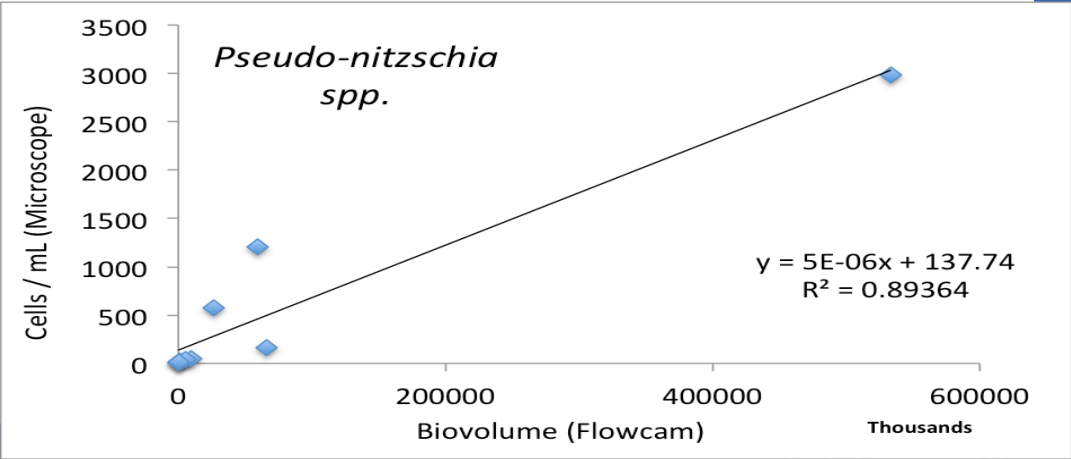
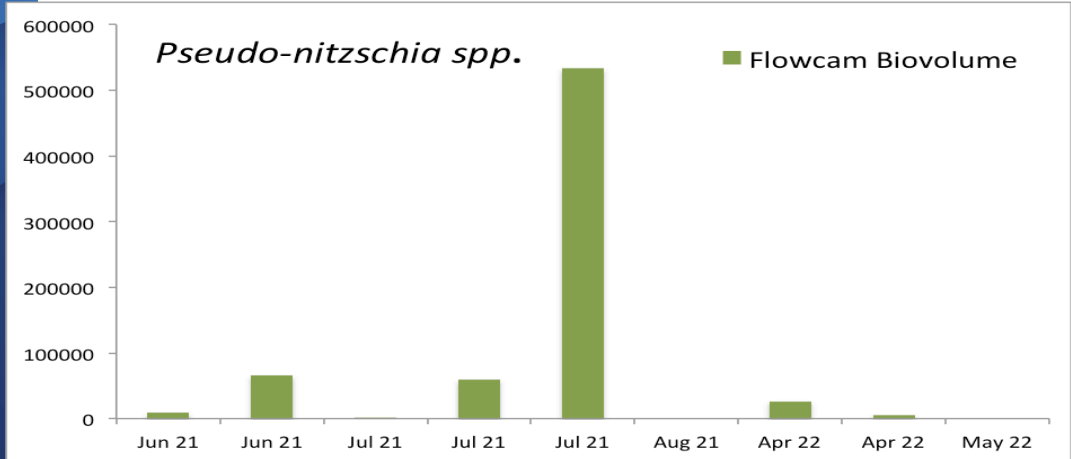
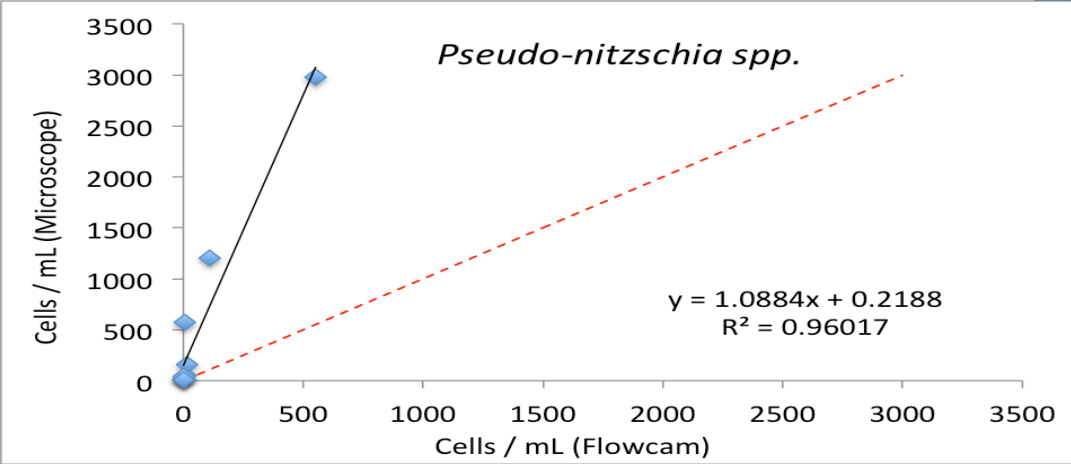
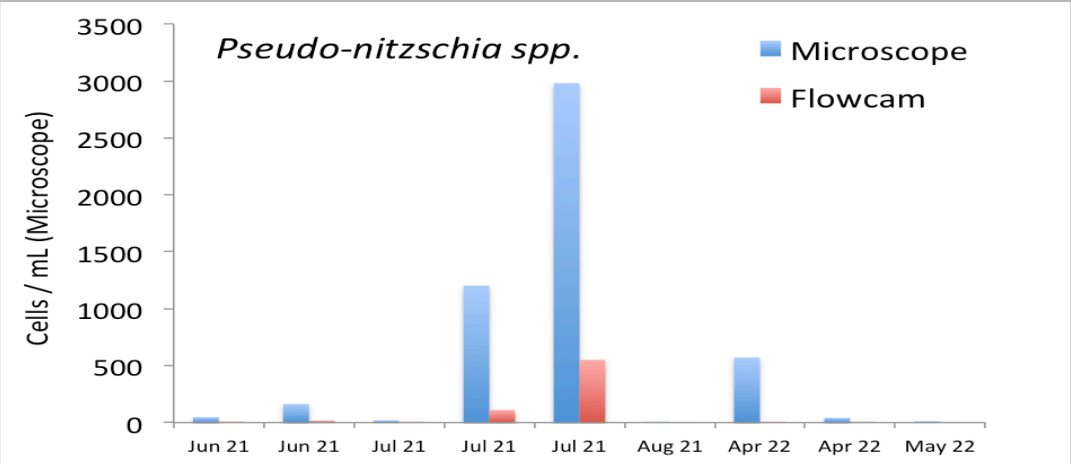
Microscope-Flowcam comparisons



Microscope-Flowcam comparisons



Microscope-Flowcam comparisons



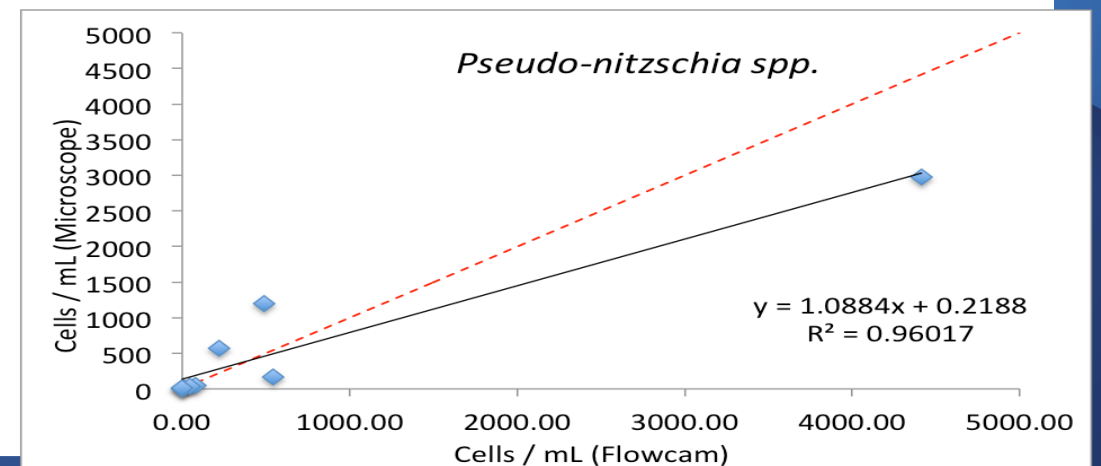
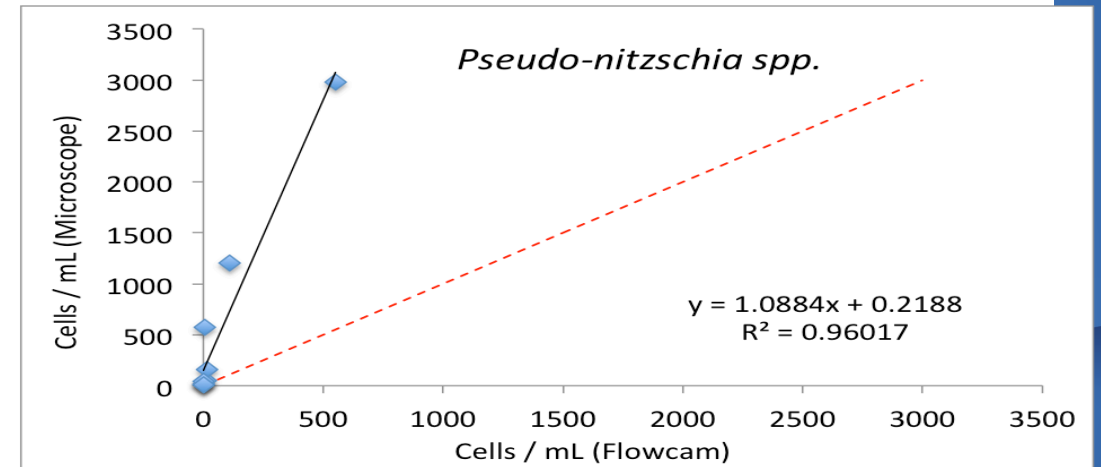
Microscope-Flowcam comparisons

Challenge of quickly enumerating chained species:

Use biovolume or biomass estimates

Classify different chain lengths

Estimate abundance from ESD volume of single cells and cumulative biovolume



Cost-effort analysis for Flowcam (4x) 10mL

Factor	Microscope	Flowcam
Preparation time	24-48 hrs / sample	0
Count time	2-4 hrs / sample	10 mins / sample
Classification time	0	20 mins / sample
Cost	8000-16000 DKK 1000-2000 USD /Euros	2000 DKK 270 USD / Euros
Data Delivery	1-2 weeks	< 1 day

Thank you!

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 - Ian Salter, ians@hav.fo

