

# **GlobalHAB Symposium 2022**

**Kristineberg Centre for Marine Research**

**22/08/2022 – 26/08/2022**

**Rob Lievaart & Vikram Rao**





# *Who are we?*

CytoBuoy BV. - A Dutch company with over 25 years of experience in manufacturing particle analysis instruments and accessories





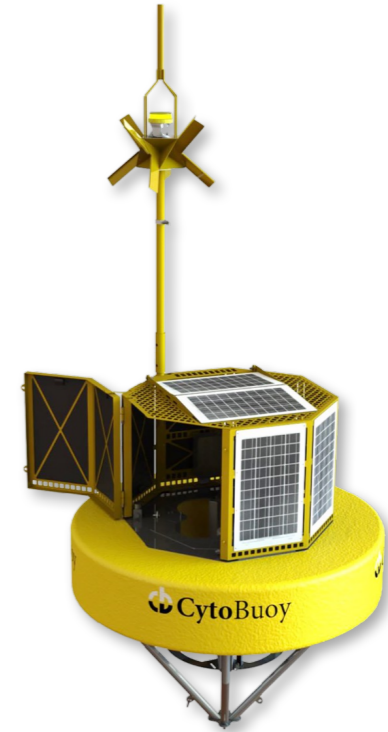
# *Main Products*



CytoSense



CytoSub



Buoy



# CytoSense



- Benchtop version of the imaging flow cytometer
- On board a ship in a ferry box setting
- Easily connected to accessories for increased automation
- Internal or external additional filtration, beads and biocide system
- 1 Laser or 2 Lasers  
(405,445,473,448,532,552,561,594,637)
- 1FWS, 2 SWS, 6 fluorescence detectors
- Anti-shock frame withstanding vibration (handy on board a ship)
- Remotely adjustable injector system
- Anti-clogging system
- No pre-filtration required



# CytoSub



- All of the features of the CytoSense
- Depth ranging 2m, 20m and 200m max.
- On a buoy, platform or mooring
- Modified into a laboratory setting
- Internal filtration, beads and biocide system (left unattended and remotely operated)
- Remotely adjustable injector system



# Buoy



- 4 compartments in the frame
- Solar Panels
- Argos system
- Telemetry System
- Battery Pack



# Features

		CONDITION	VALUE	ENTITY	REMARKS
<i>Particle size &amp; sample size</i>	smallest particle size	equal or smaller	0.1 / 0.4	µm	Detection: Sideward scatter. Depending on the refractive index (For example: 0.1 for polystyrene microspheres, 0.4 for cells like Prochl.m.)
	minimum system orifice dimension	equal or bigger	800	µm	the minimum system orifice limits the size of organisms that can flow through
	maximum analyzed particle length	equal or longer	2.500	µm	this allows filamentous and elongated organisms to be analyzed correctly
	minimum volumetric sample flow rate	equal or smaller	5	µl/min.	the low flow rate is important to analyse samples with small particles at high abundance without dilution
	maximum volumetric sample flow rate	equal or bigger	1.000	µl/min.	the high flow rate is important to analyse samples with low particle concentration
	particle concentration		$10^3 - 10^{11}$	particles/L	$10^{11}$ means 7,000 particles per second @ ca. 10% coincidence

		CONDITION	VALUE	ENTITY	REMARKS
<i>Data format</i>	full signal pulse profiles (scans) per particle	meet	available		allows morphological analysis of larger organisms, linear biomass determination and high sensitivity
	maximum particle analysis (scan) rate	equal or bigger	10.000	particles/s	the count rate is important to analyse samples with smaller cells at high abundance (increases coincidence)
	maximum particle scans	equal or more	1.250.000	particles	maximum amount of particle scans per file



# Features

		CONDITION	VALUE	ENTITY	REMARKS
<i>Particle imaging</i>  <i>optional</i>	maximum imaging rate	equal or more	20	photos/s	
	maximum photos per file	equal or more	10.000	photos	
	optical photo resolution	equal or smaller	0.8	µm	resolution defined by the quality of the optics. NA=0.4.
	digital photo resolution	equal or more	3.3 4.6*	pixels/µm	resolution defined by the sensor * optional - high resolution camera
	image frame size (W x H)	equal or more	576 x 360 778 x 614* 528 x 441**	µm x µm	this is the size of the field of view of the camera * optional - larger sensor ** optional - higher resolution camera
	Image resolution (W x H)	meet	1920 x 1200 2448 x 2048*	pixels	* optional - higher resolution camera
	magnification	equal or more	16x	factor	
	image matching with optical signal scan	meet	yes		each image is combined with the optical signal profiles for that particle
	photo triggering based on all optical signals	meet	yes		targeted imaging: smart preselection of photos based on various combinations of the optical signals





# Operation and Software

CytoUSB5

MeasurementsWindow

GlobalHab\PhaeocystusGlobosa

Measuring

25/08/2022 22:10:29

0.52 MB

24

37 / 300s

2776 particles

37.4 ul

69.9E0 p/ul

x

37.4 ul

GlobalHab\106\_Pico\_Flr12\_5mu\_5\_min

GlobalHab\106\_Micro\_Flr25\_13mu\_20min 3/4

GlobalHab\106\_Photo\_Flr25\_5mu\_20min

GlobalHab\106\_Photo\_Large\_Flr25\_5mu\_5min

Imaging in Flow

Channels Mode Log Smoothed

[mV]

117.15

87.86

58.57

29.28

Sample length

[um]

SWS HS - Length

1e3

1e2

1e1

1e0

1e0

1e1

1e2

1e3

SWS HS

SWS LS

FL Yellow HS

FL Yellow LS

Fl Orange

Sample pump: inwards: 1.01  $\mu$ L/s

Sheath pump: 68.6%

Switches

Laser 1

Main Power

Grabber Power

+12V Preamp

Laser 2

Sheath Pump

Sample Pump

High Voltage

Blower

Pinch valve Bypass

Image in Flow

Beads

Injector

Image in Flow

Start Camera Focus Mode

Target FWS Ratio: 1

Range Width(%): 20

49 (9.4x)

47 (6.6x)

94 (254x)

76 (410x)

106 (716x)

Acquiring sample

CytoUSB

Type here to search

23°C Meest bewolkt

22:11

25/08/2022



# Operation and Software

CytoClus4

File Cytoqram Other Plots

Database

Example: "beads"

Folder

O... File name Start

GlobalHab (57 items)

- 106\_Micro\_Flr25\_13mu\_5... 24/08/2022
- 106\_Photo\_Large\_Flr25\_5... 24/08/2022
- 106\_Photo\_Flr22\_5mu\_20... 24/08/2022
- 106\_Micro\_Flr22\_13mu\_2... 24/08/2022
- 106\_Pico\_Flr12\_5mu\_5\_mi... 24/08/2022
- All 2022-08-24 11h28.cyz 24/08/2022
- All 2022-08-24 11h09.cyz 24/08/2022
- Beads 2022-08-23 20h42... 23/08/2022
- Beads 2022-08-23 20h31... 23/08/2022

Database Set statistics

Set Library

View Name

- Default (all)
- Rhodomonas
- All Imaged Particles
- HeteroSigmaAkishiwo
- KartodiniumVentricum
- Chatonella
- KarenijaMikim
- Alexandrium
- Melosina
- PhaeocystusGlobosa
- PrymniusKarvum
- P\_Polylepis
- Pyromonas
- ChrysoChromati
- Unassigned Particles

Exclusive Sets

01: 106\_Micro\_Flr25\_13mu\_20min 2022-08-24 21h51 02: All 2022-08-24 11h09

Particle # 8938

Log Channels

Level [mV]

Signal Length (μm)

FWS Length: 153.4 μm  
SWS HS Length: 170.5 μm  
FL Red HS Total: 134294.5

Image Overview - Default (all)

36% Maximum Image Size:

219 μm

Mouse instructions  
Left: drag the image  
Middle: draw measure tool  
Scroll: zoom  
Right: reset zoom

Page 1 Page 2

Total FL Red HS - Total FL Yellow HS

Mouse: Gates: Move

Total FL Yellow HS ( $\mu = 429.75, \sigma = 3735.29$ )

Total FL Red HS ( $\mu = 1732.55, \sigma = 5692.11$ )

Maximum FL Red HS - Maximum FL Ye

Mouse: Gates: Move

Maximum [mV] FL Yellow HS ( $\mu = 36.81, \sigma = 283.76$ )

Maximum [mV] FL Red HS ( $\mu = 138.08, \sigma = 308.8$ )

Alignment Plot

Mouse: Gates: Move

Default (all): 29,32% left biased - 70,68% right biased  
73,28% within margin

Total FWS R ( $\mu = 2332.92, \sigma = 7919.43$ )

Total FWS L ( $\mu = 1941.07, \sigma = 7370.74$ )

Lower tolerance: 0,50 Upper tolerance: 0,50

Total FI Orange - Total FL Yellow HS

Mouse: Gates: Move Clear

Total FL Yellow HS ( $\mu = 429.75, \sigma = 3735.29$ )

Total FI Orange ( $\mu = 371.80, \sigma = 3144.02$ )

Total FL Red LS - Total FL Yellow HS

Mouse: Gates: Move

Total FL Yellow HS ( $\mu = 429.75, \sigma = 3735.29$ )

Total FL Red LS ( $\mu = 17.08, \sigma = 83.10$ )

CS-2021-106 - Lobith

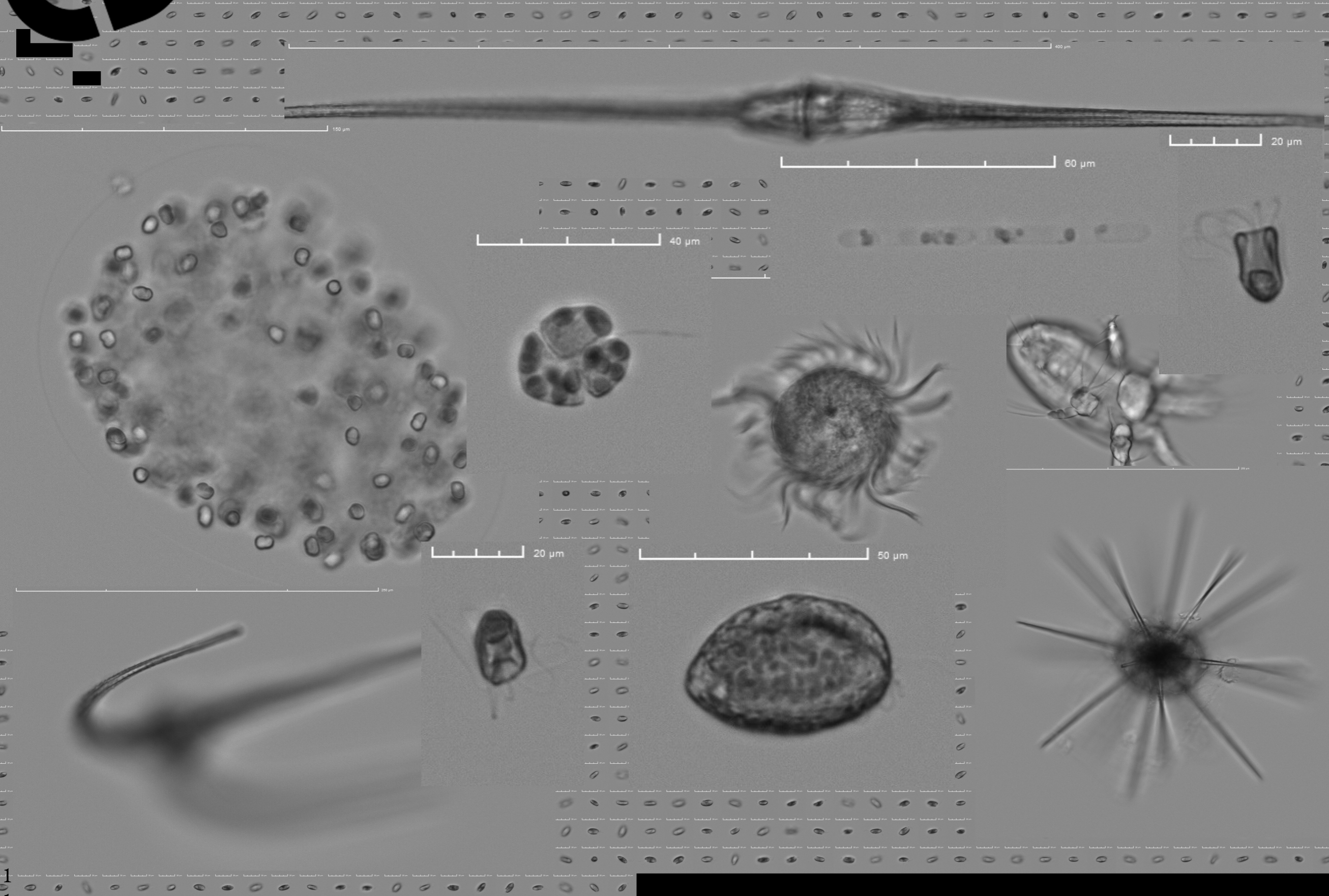
Memory usage: 5,146 GB

Type here to search

20°C Molnigt 20:58 25/08/2022



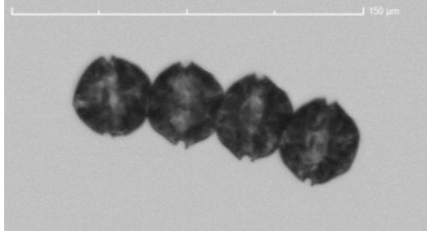
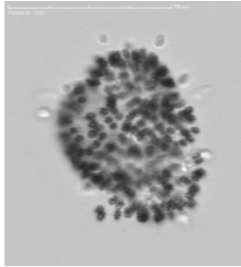
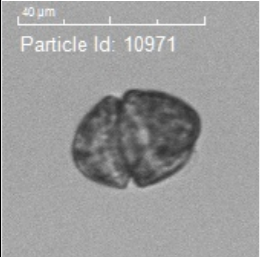
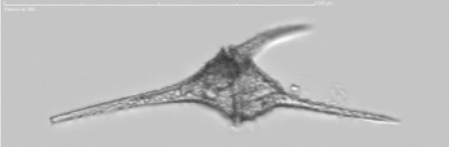
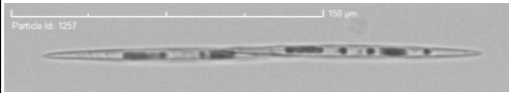
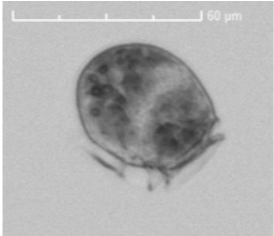

# Images





# Images

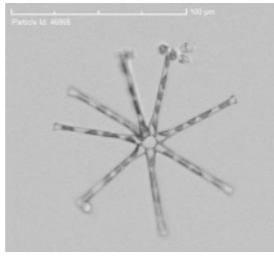

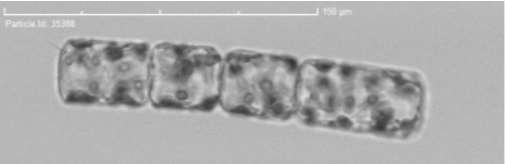
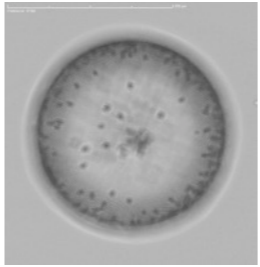
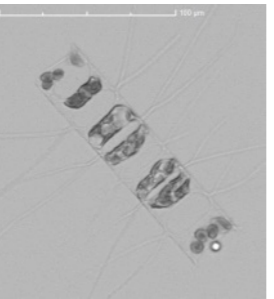
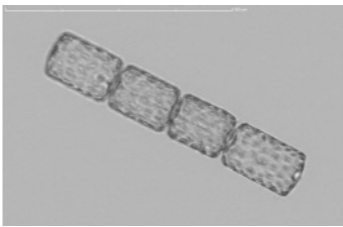
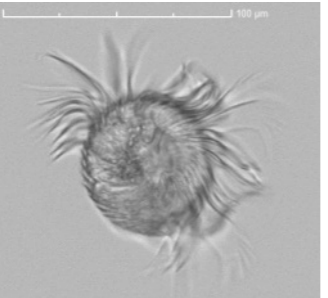
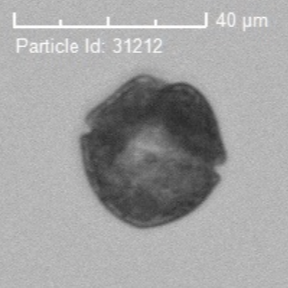

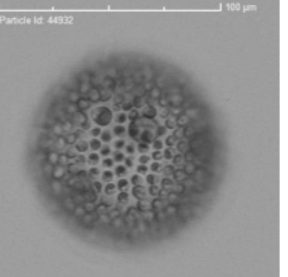
## Some HAB species detected by CytoSense

	<i>Alexandrium</i>		<i>Microcystis</i>
	<i>Gymnodinium</i>		<i>Ceratium</i>
	<i>Pseudo-nitzschia</i>		<i>Dinophysis</i>
	<i>Prorocentrum</i>		



# Images

## Other species detected by CytoSense

	<i>Asterionella</i>		<i>Coleps</i>
	<i>Cetrataulina</i>		<i>Coscinodiscus</i>
	<i>Chaetoceros</i>		<i>Dactyliosolen</i>
	<i>Ciliates</i>		<i>Dinoflagellates</i>
	<i>Zooplankton</i>		<i>Volvox</i>



# Accessories



Sampling Automation: A lab system enables you to monitor multiple individual sample stations with individual scheduling



Staining Module: A device for fluorescent staining, coupled to a CytoSense, for automated online analysis of microbial groups of autotrophs (algae), mixo- and heterotrophs (bacteria, microzooplankton) and other particles



Automated Filtration System: The recirculating sheath fluid is kept clean and clear for long periods by an autonomy system consisting of filters, biocide dosing, activated carbon and calibration diagnostics.



# Partners



**izasa**  
**scientific**  
a werfen company

## Our partners





# *New Product*



**CytoSense XR**





Thank You