



Fluorescence Activated Cell Sorting (FACS)

as a tool for rapid establishment of microalgal cultures at
the National Bank of Algae, Canary Islands, Spain

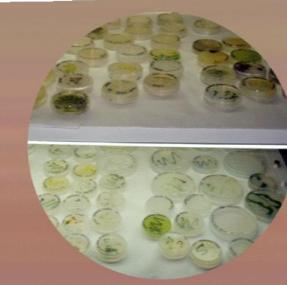
Paulo S. Salomon, Ana M. Giraldo, Guillermo G. Reina

National Bank of Algae (Banco Nacional de Algas – BNA)

Founded in 1998
Part of the Marine Biotechnology Center - ULPGC

International
Depository Authority for microorganisms recognized by
WIPO (World Intellectual Property Organization) in 2005

Member: ECCO since 2001
WFCC since 2003

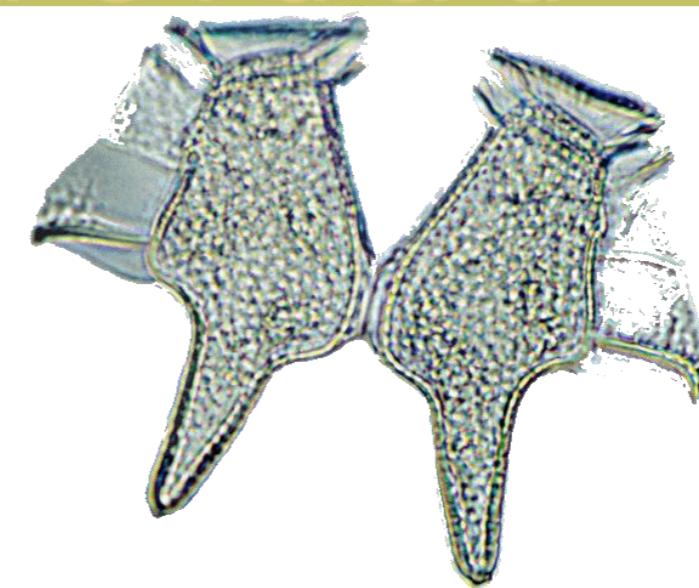
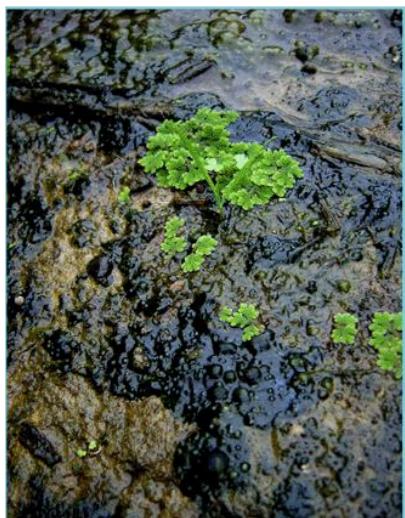




Isolation and preservation of cyanoprokaryota and microalgae from the Macaronesic area and the African coast nearby, with special interest in threatened species or those which grow in special environments.



Biodiversidad





Macaronesia

Azores

Madeira

Savage

Canary Islands

Cabo Verde

The image shows a satellite view of the North Atlantic Ocean, focusing on the Macaronesia region. The region includes several island groups: the Azores (a chain of islands in the central Atlantic), the Madeira Islands (a small group off the coast of Portugal), the Savage Islands (a small group near the Azores), the Canary Islands (a large group off the coast of Africa), and Cabo Verde (a group further west). The landmasses are shown in various shades of green and brown, indicating different vegetation and terrain. The surrounding ocean is a deep blue, with some white foam visible at the edges of the islands.



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The Canary Islands

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2009 → 2.6 million€

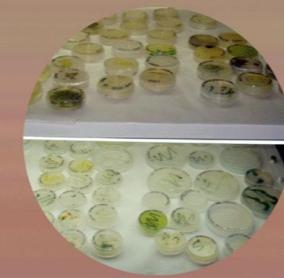


GOBIERNO
DE ESPAÑA

MINISTERIO
DE CIENCIA
E INNOVACIÓN

National Strategic Plan for Microalgae Isolation and cultivation of microalgae for:

- Carbon Sequestration
- Biofuel production





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Big PBR

Islandia90_023

Gulfos Arroyo rojo



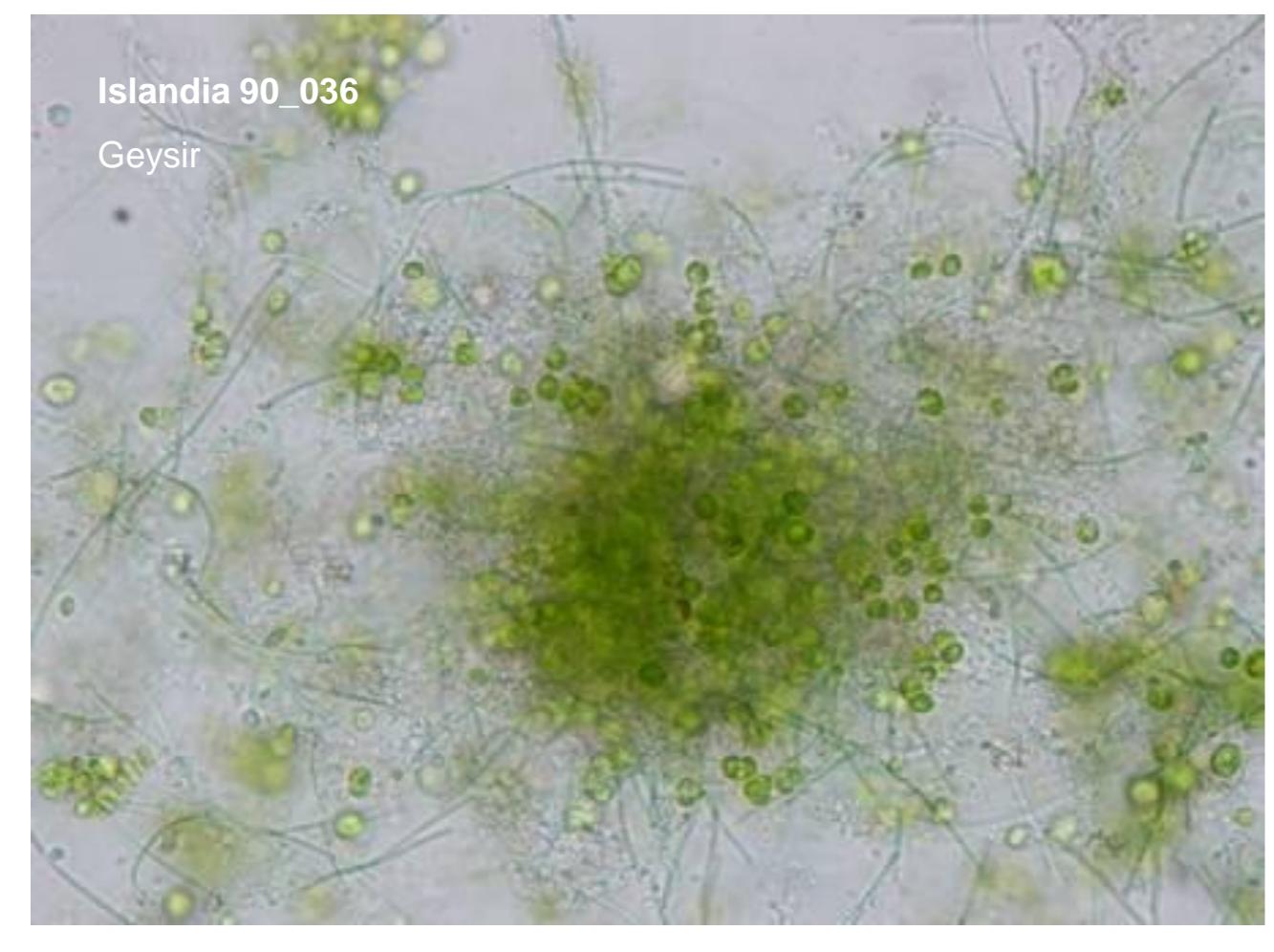
Fuerteventura

Tapete Gorriones



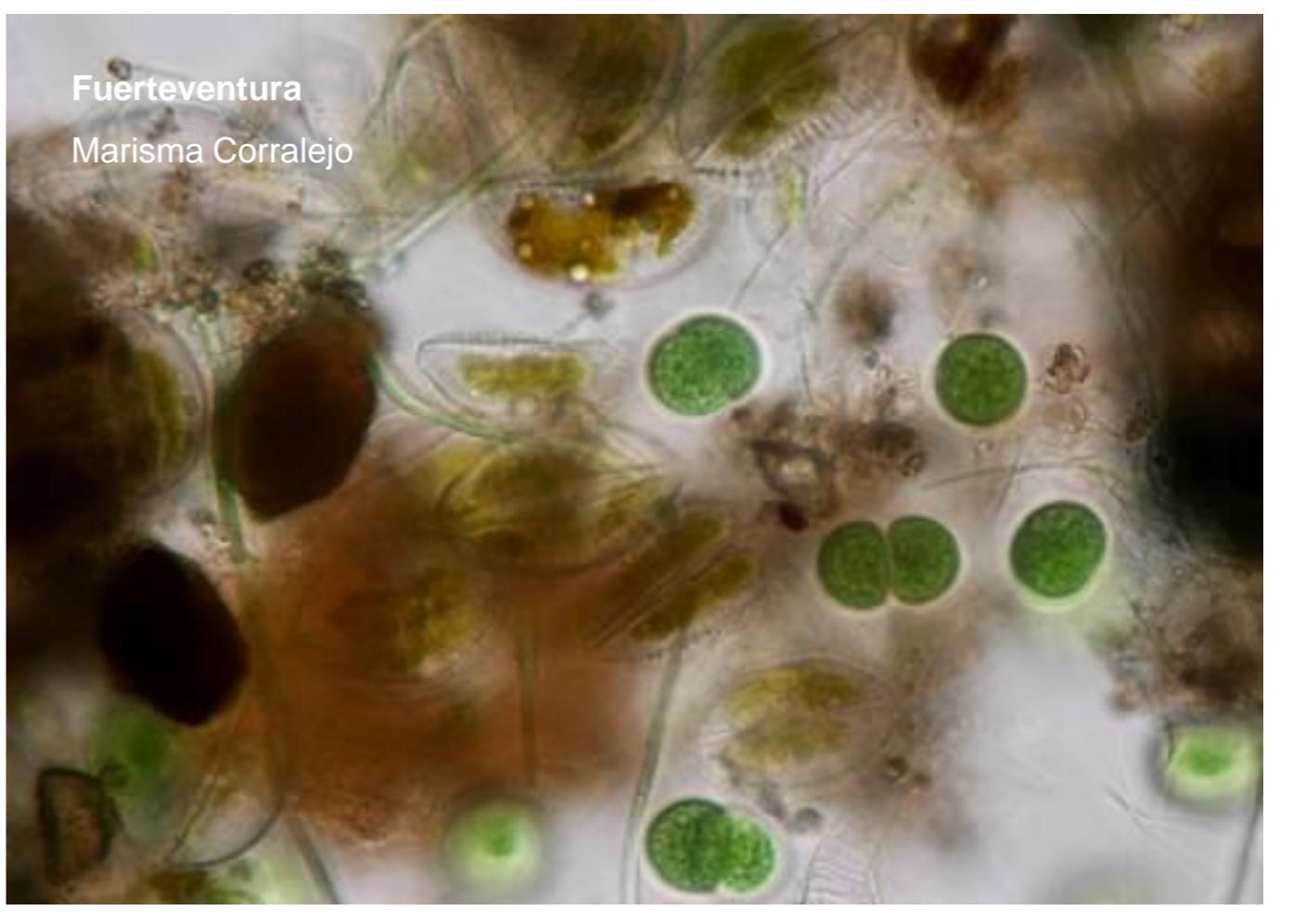
Islandia 90_036

Geysir



Fuerteventura

Marisma Corralejo





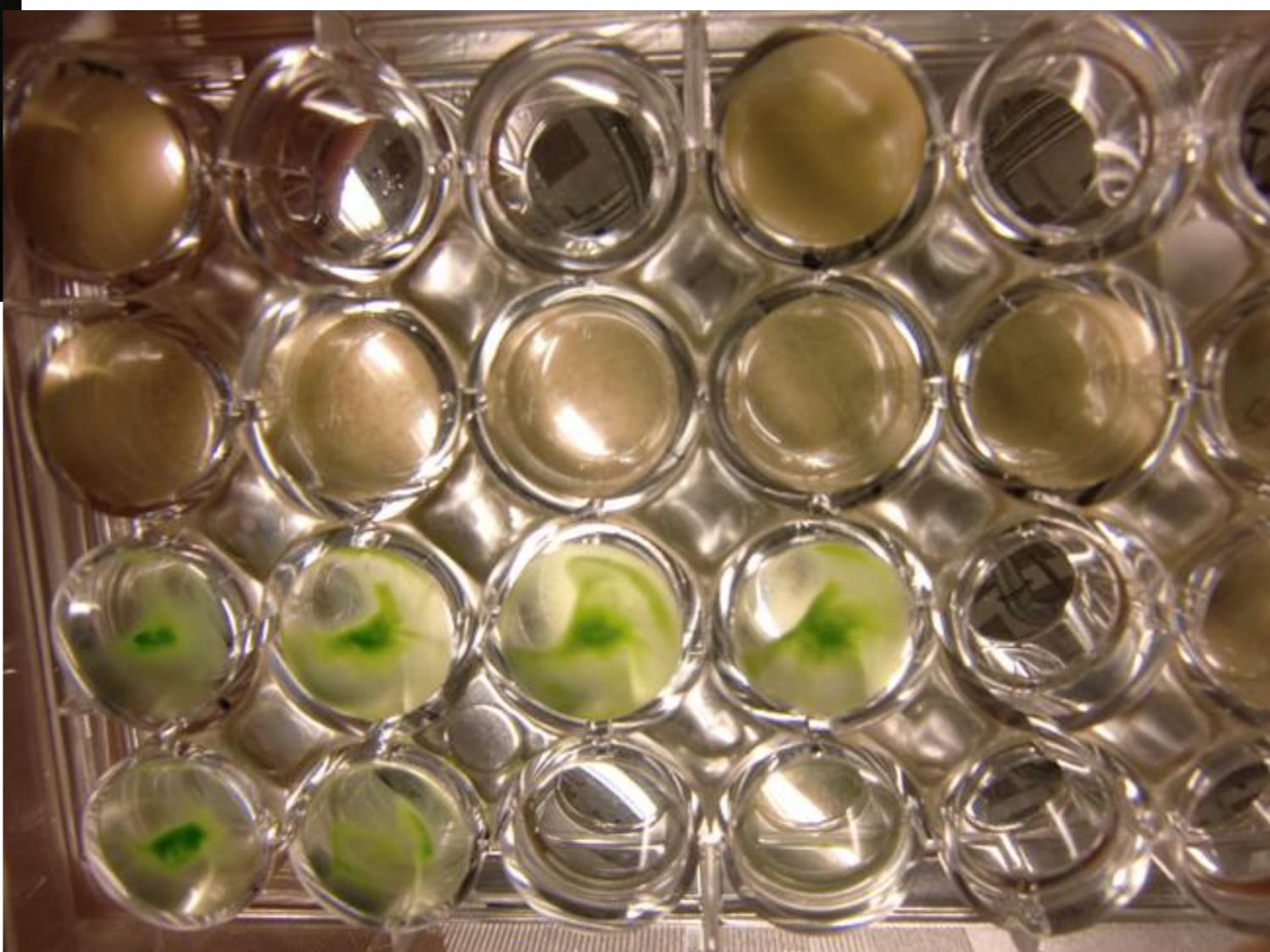
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Traditionalsolation

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Micro capillarypipette



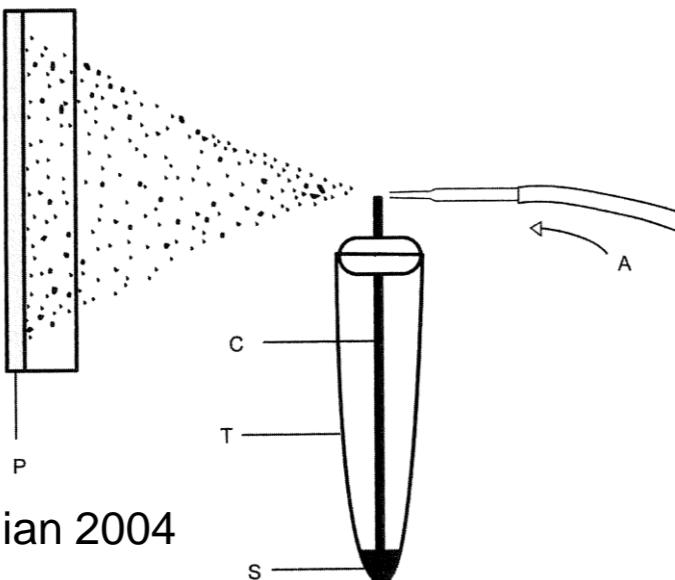


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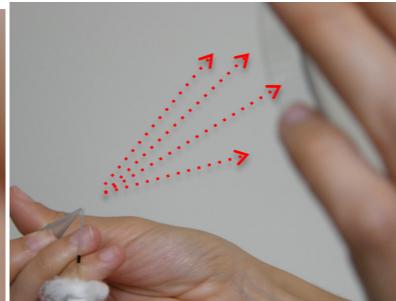
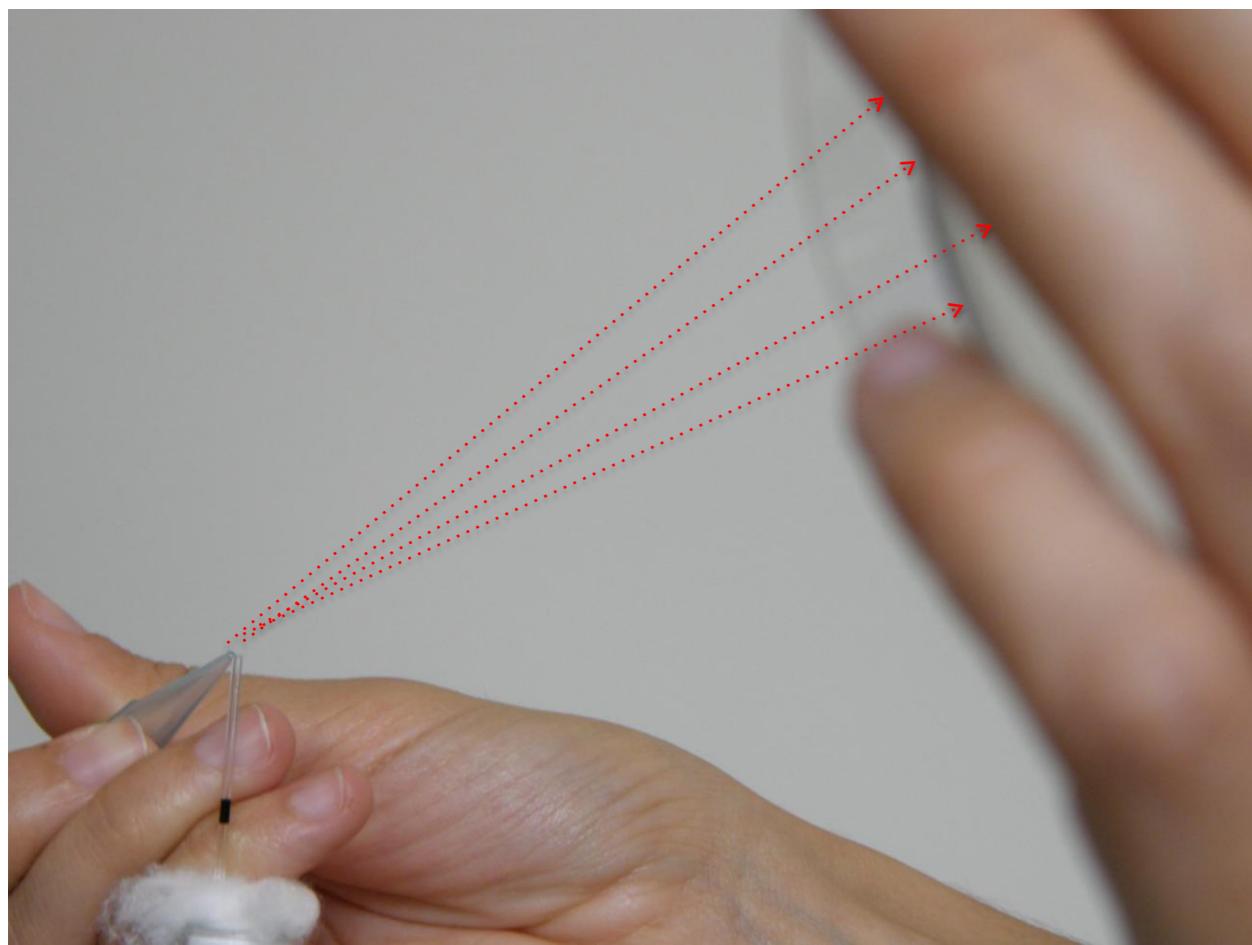
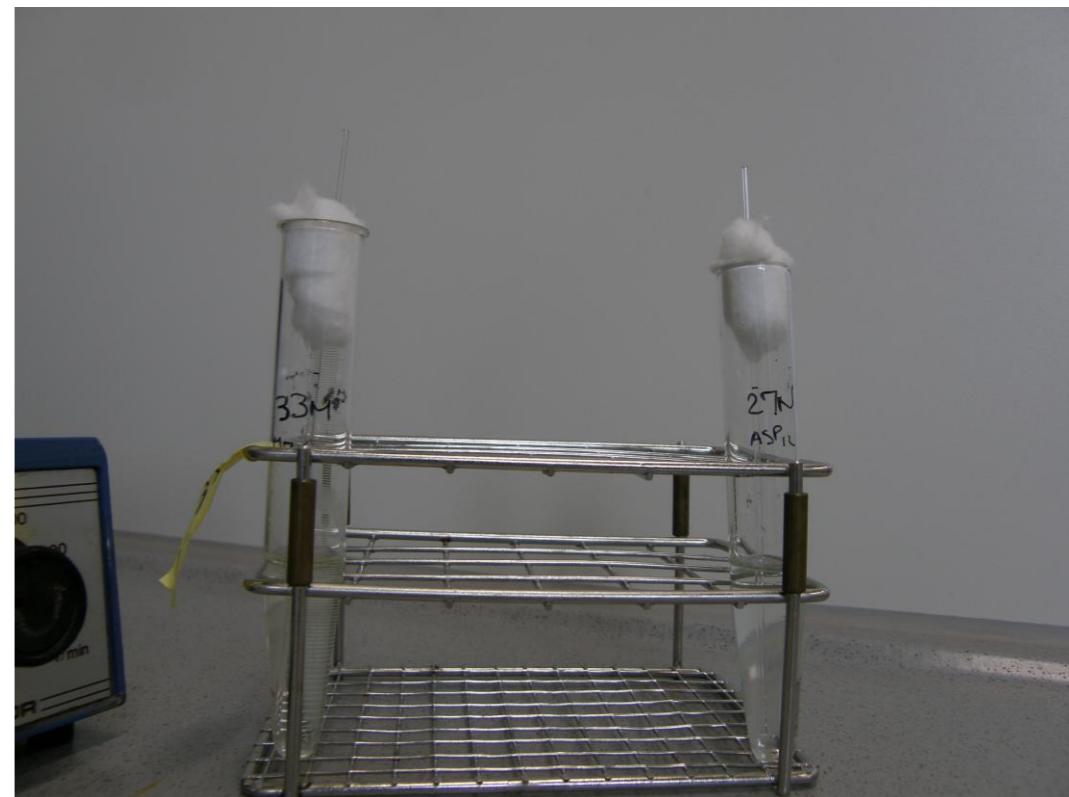
Traditionalsolation

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Sprayingon agar



Surek&Melkonian 2004

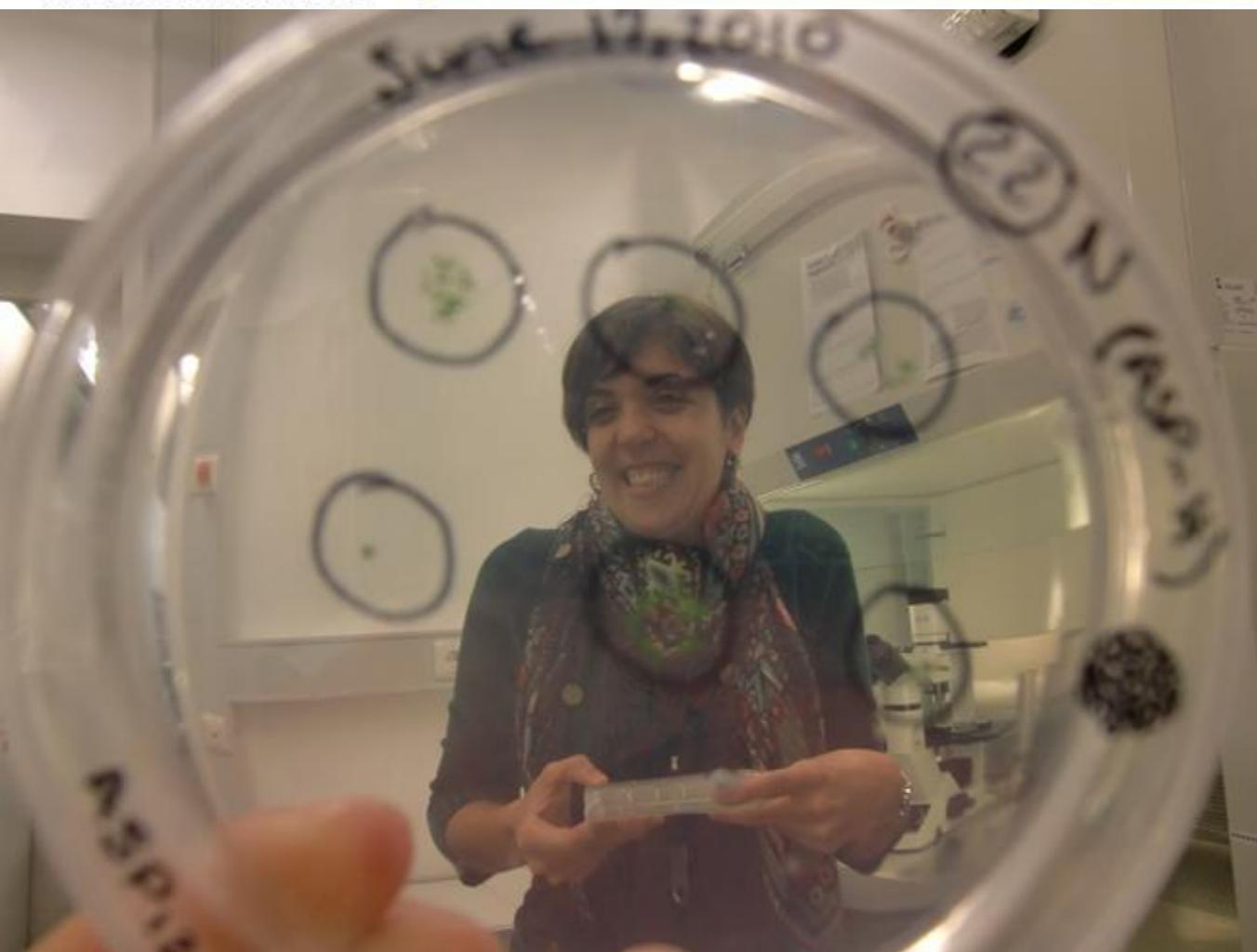




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Traditionalsolation (manualy)

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Moving cells on/through agar



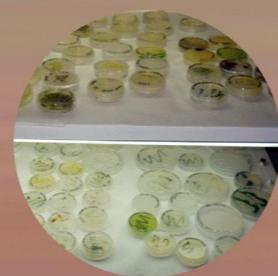
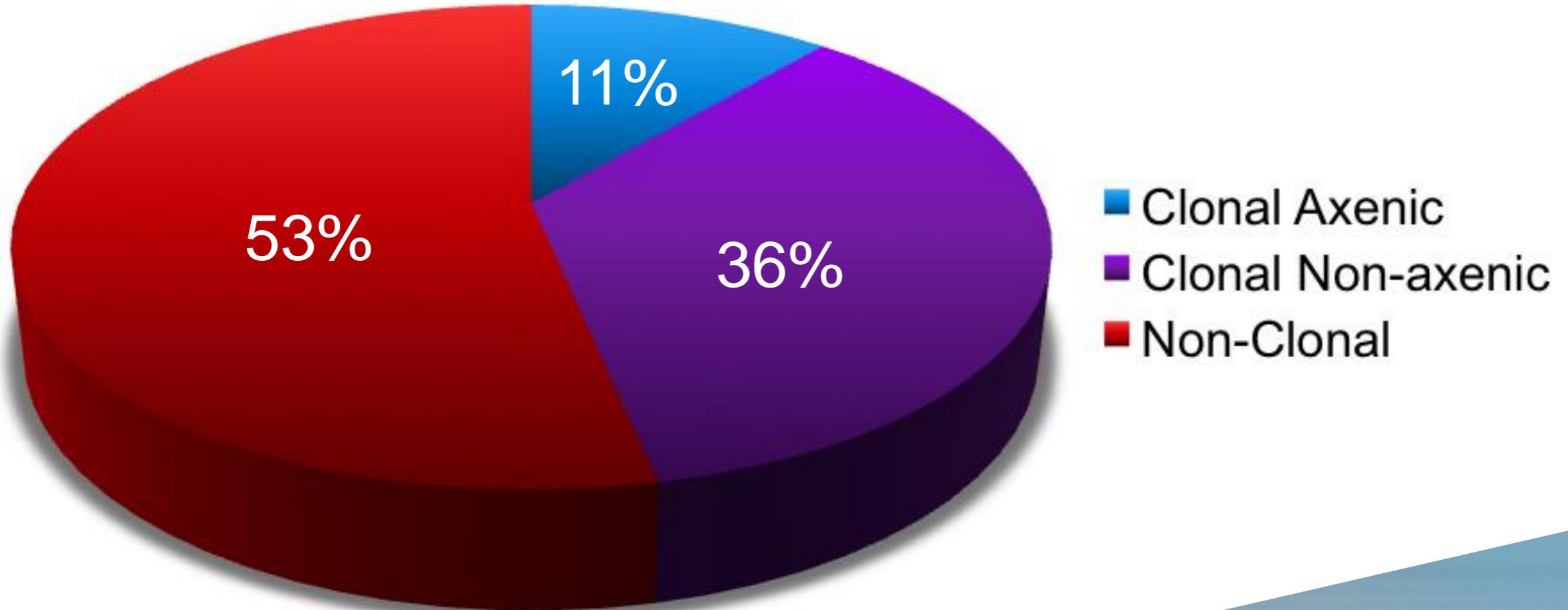


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Collection of eukaryotic microalgae and cyanobacteria

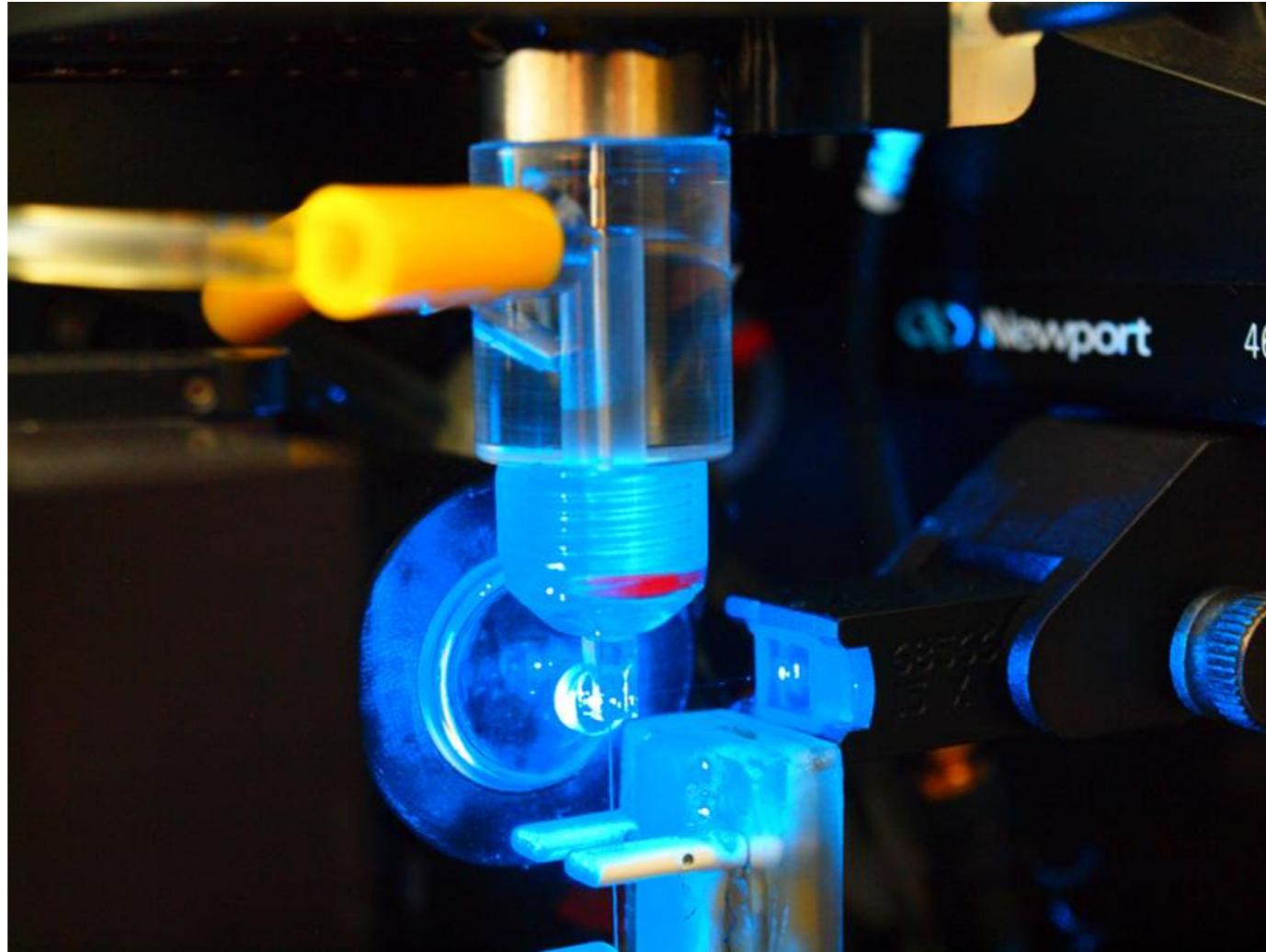
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BNAs holds 110 strains... and counting





Fluorescence-Activated Cell Sorting



FlowCytometer



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Principle of FACS

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Detection



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Principle of FACS

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Sorting



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Algal cells have pigments ...

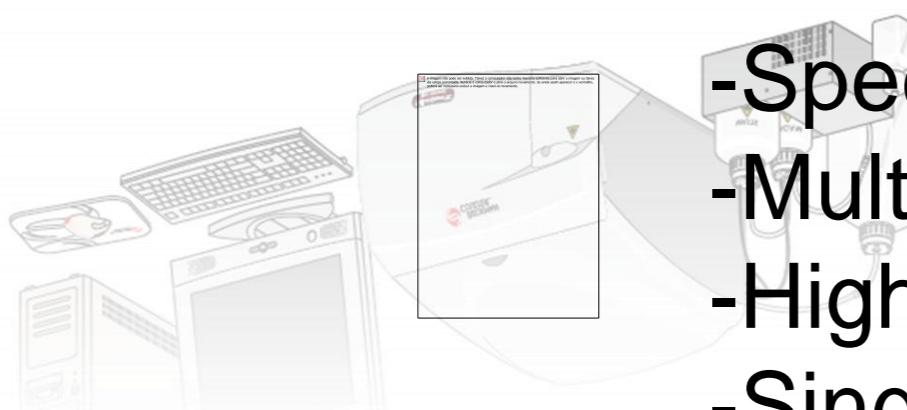
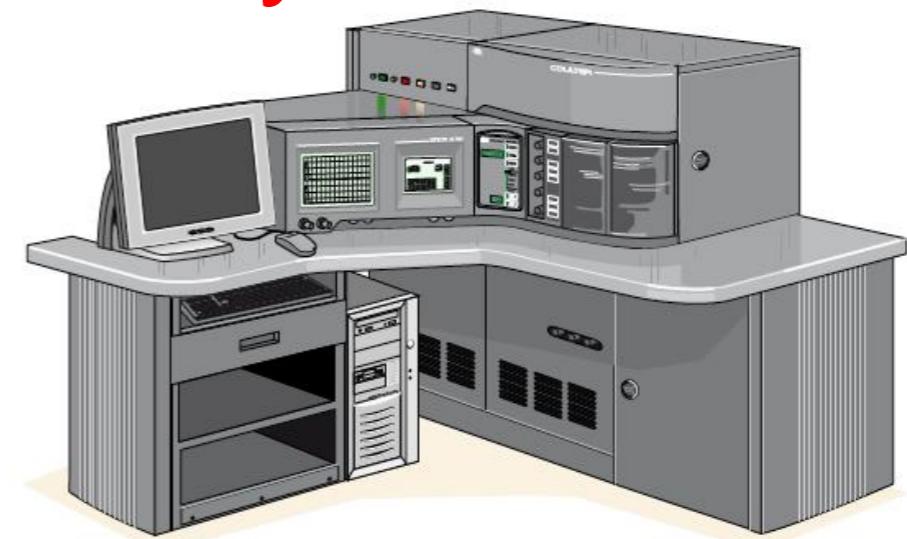
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**Detection and sorting of live cells
Production of clonal, axenic cultures in one go**

Analysis



Analysis + Sorting



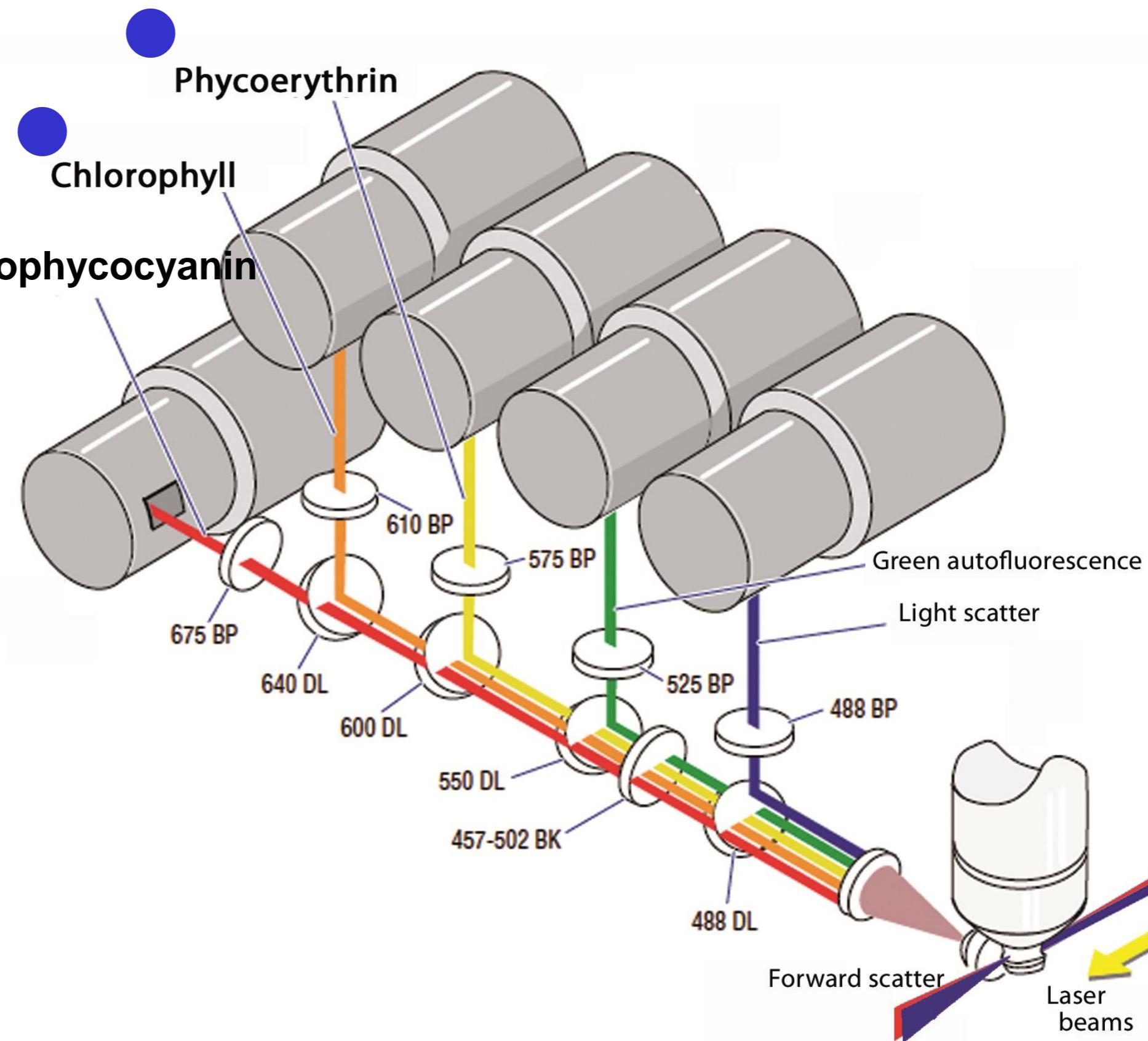
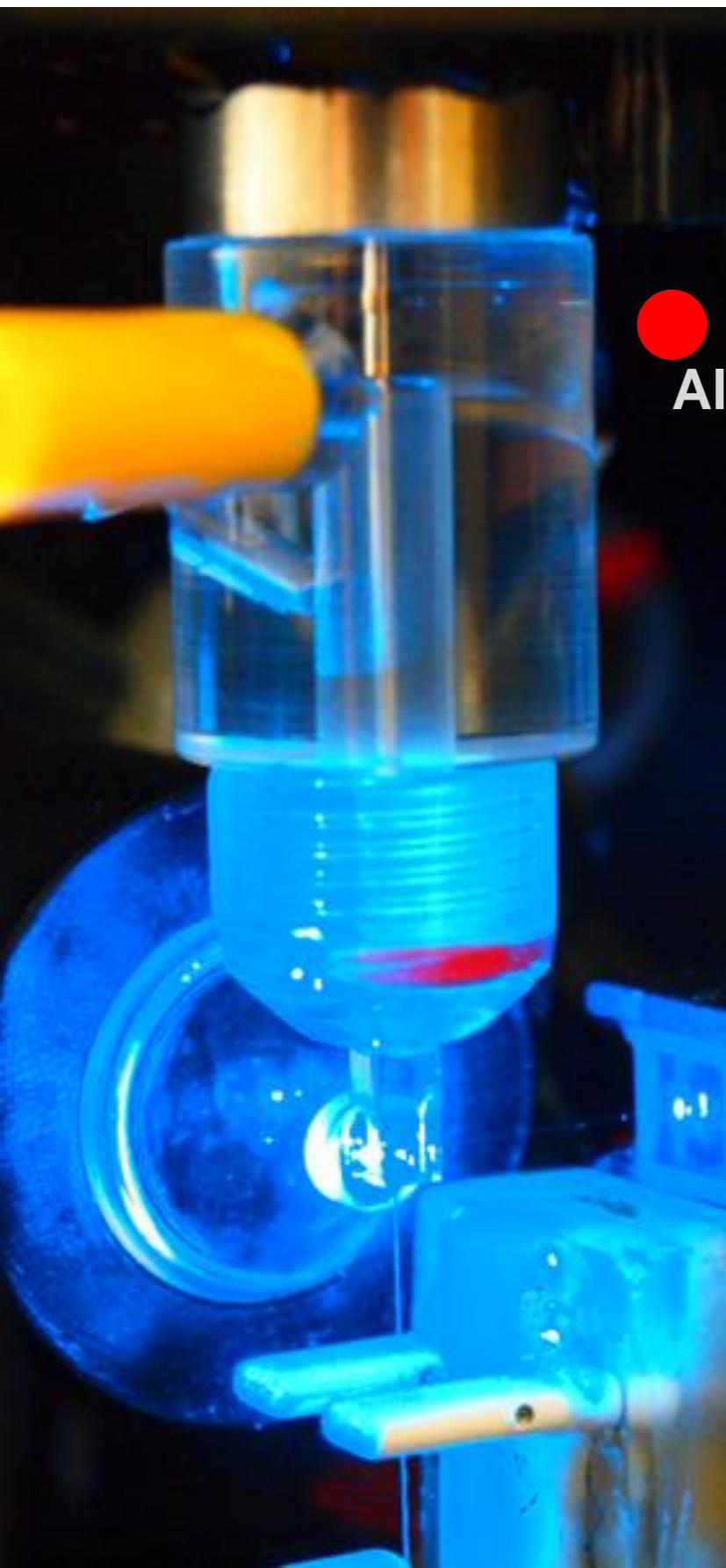
- Speed (1 000 cells s^{-1} or more)
- Multiparameter (simultaneously measured)
- High sensitivity
- Single cell analysis (one at a time)
- Measurements on high number of cells
(100 000 cells in less than 2 min, good for statistics)
- Detection of rare events



Beckman-CoulterEpics ALTRA

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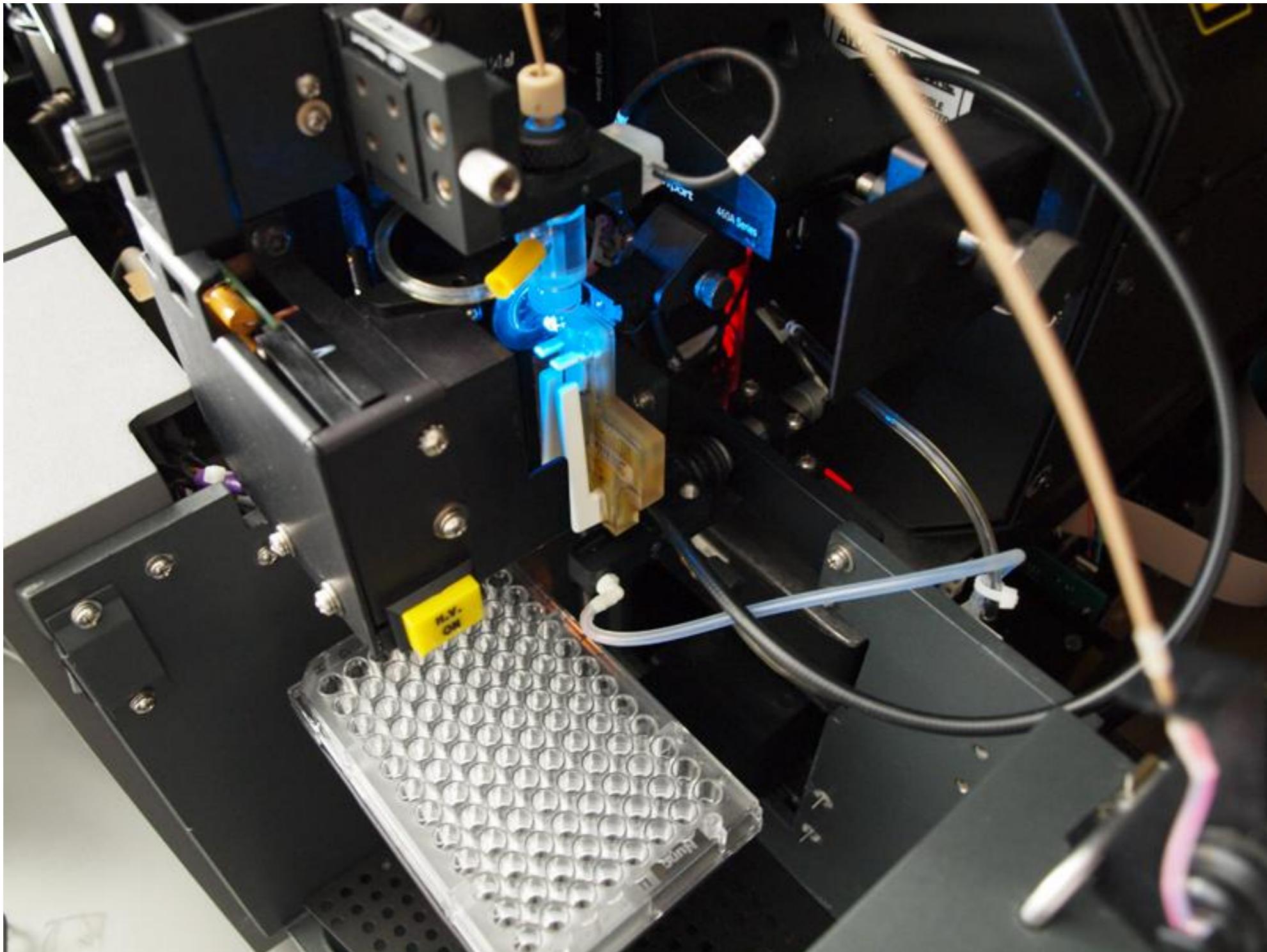


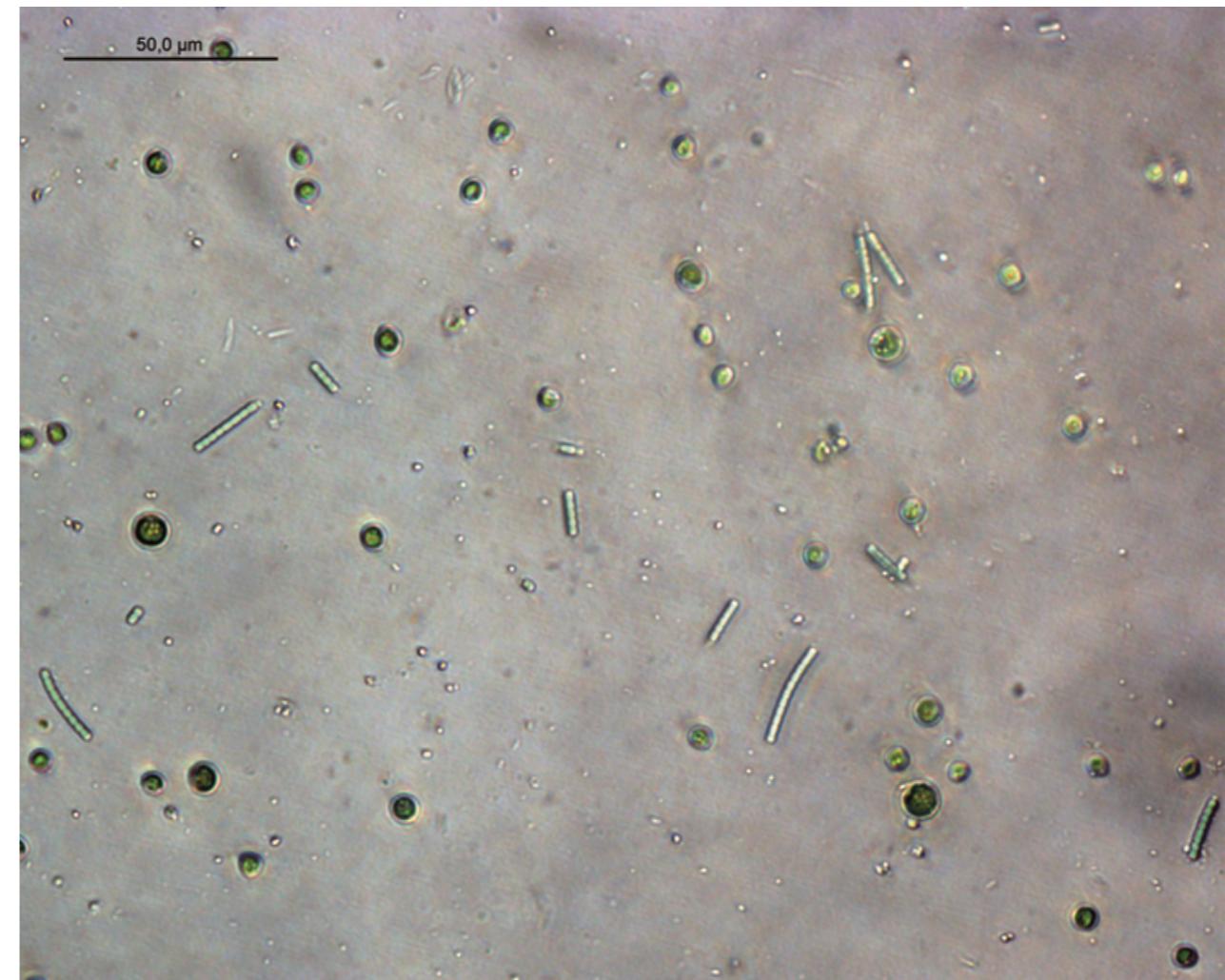
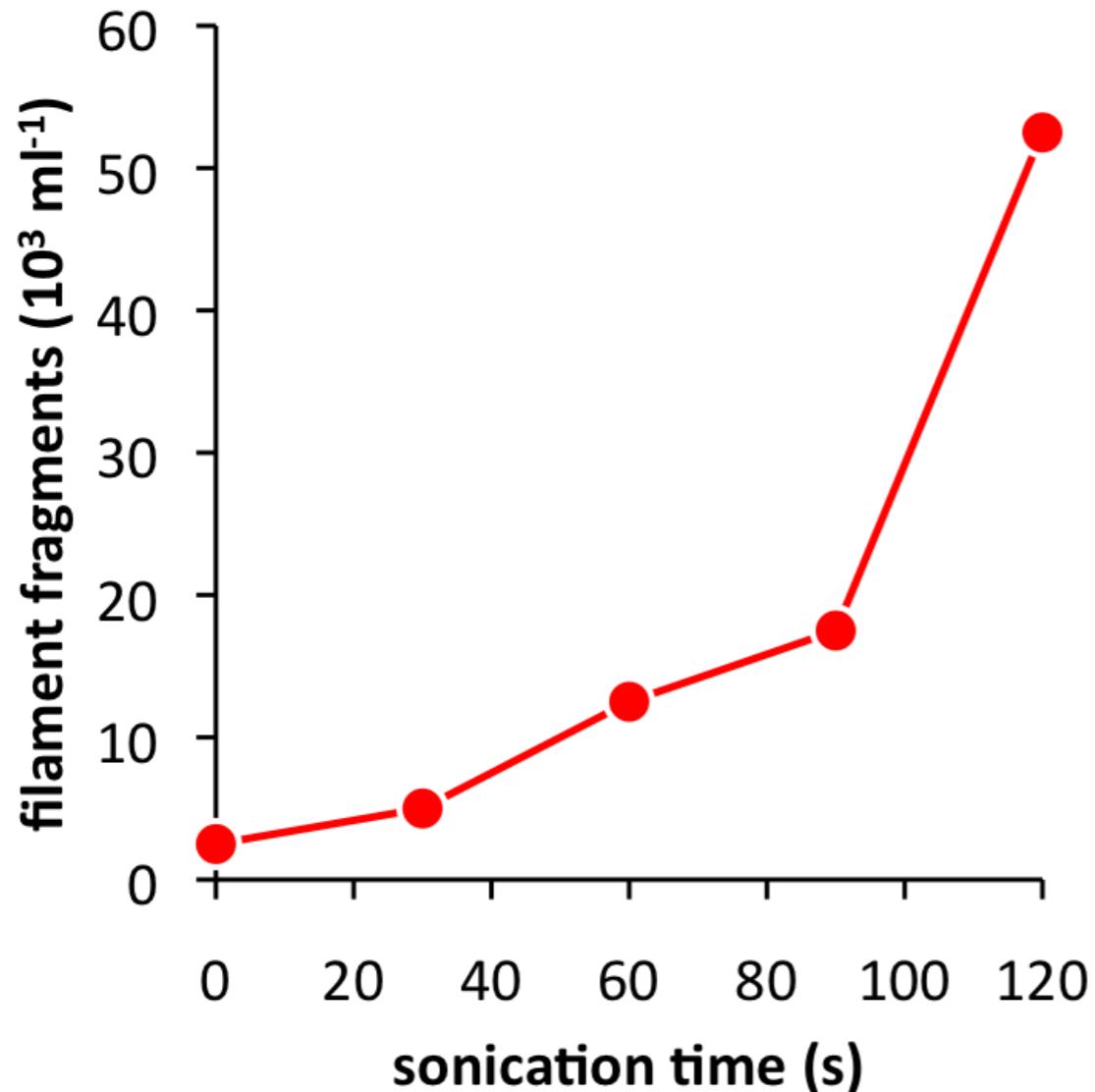


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Autocloneoption

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Do microalgal cells come out alive of the sorting process?

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- ✓ **High pressures (e.g. > 30 PSI)**
- ✓ **Acceleration into the fluidics system**
- ✓ **LASER illumination (e.g. > 150 mW)**
- ✓ **Electric charges (droplet deflection)**
- ✓ **Deceleration (impact on collection device)**

Eur. J. Phycol. (1993), **28**: 93–97. Printed in Great Britain

The production of clonal and axenic cultures of microalgae using fluorescence-activated cell sorting

**20% survival
20% axenic**

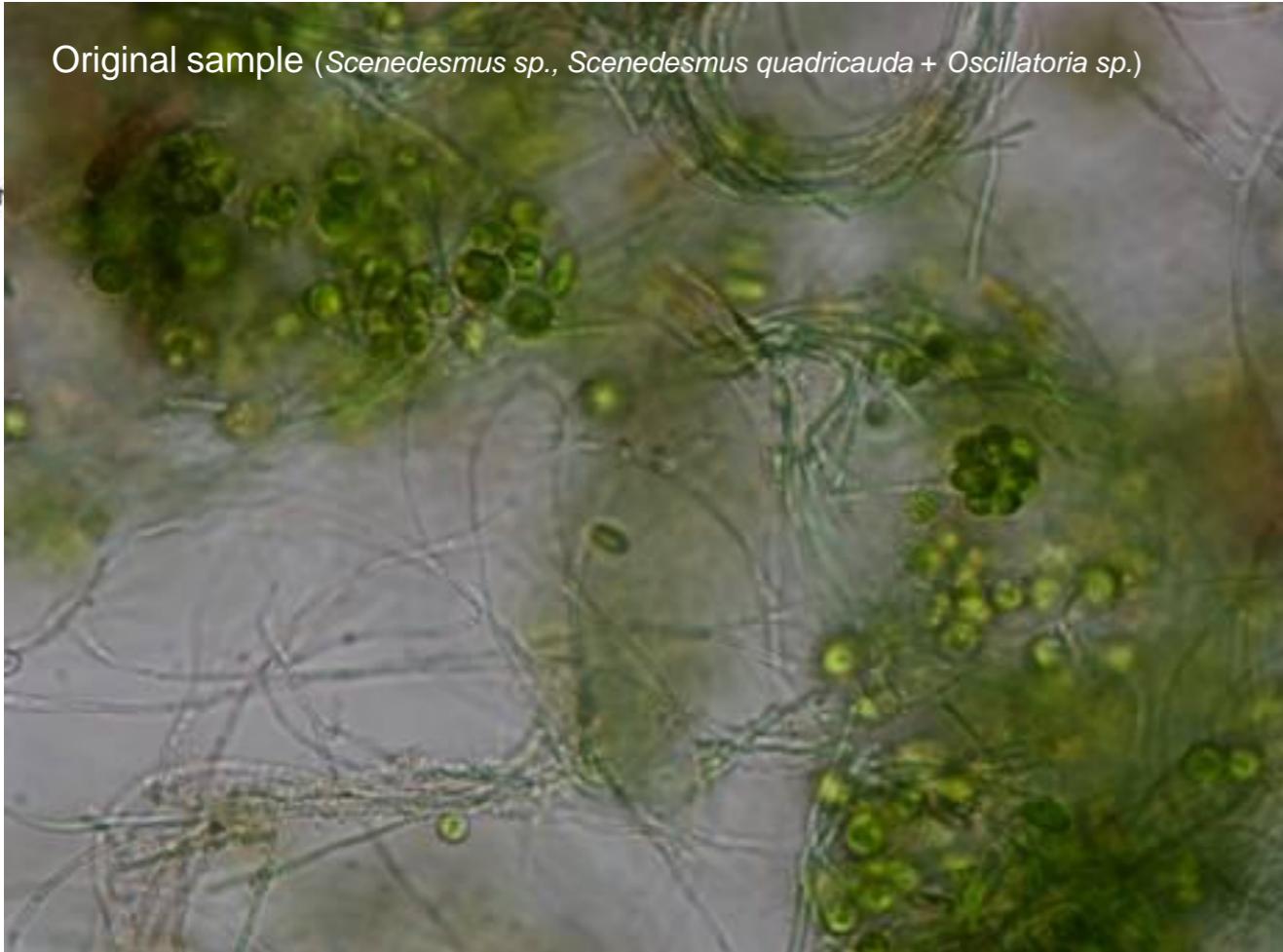
Species used by Sensen et al 2003

CHRISTOPH W. SENSEN, KIRSTEN HEIMANN AND MICHAEL MELKONIAN

Universität zu Köln, Botanisches Institut, Lehrstuhl I, Gyrhofstrasse 15, D-5000 Köln 41, Federal Republic of Germany

Sensen et al. 2003

Original sample (*Scenedesmus* sp., *Scenedesmus quadricauda* + *Oscillatoria* sp.)



Isolation of *Scenedesmus* sp. In the flow cytometer

Change in morphology

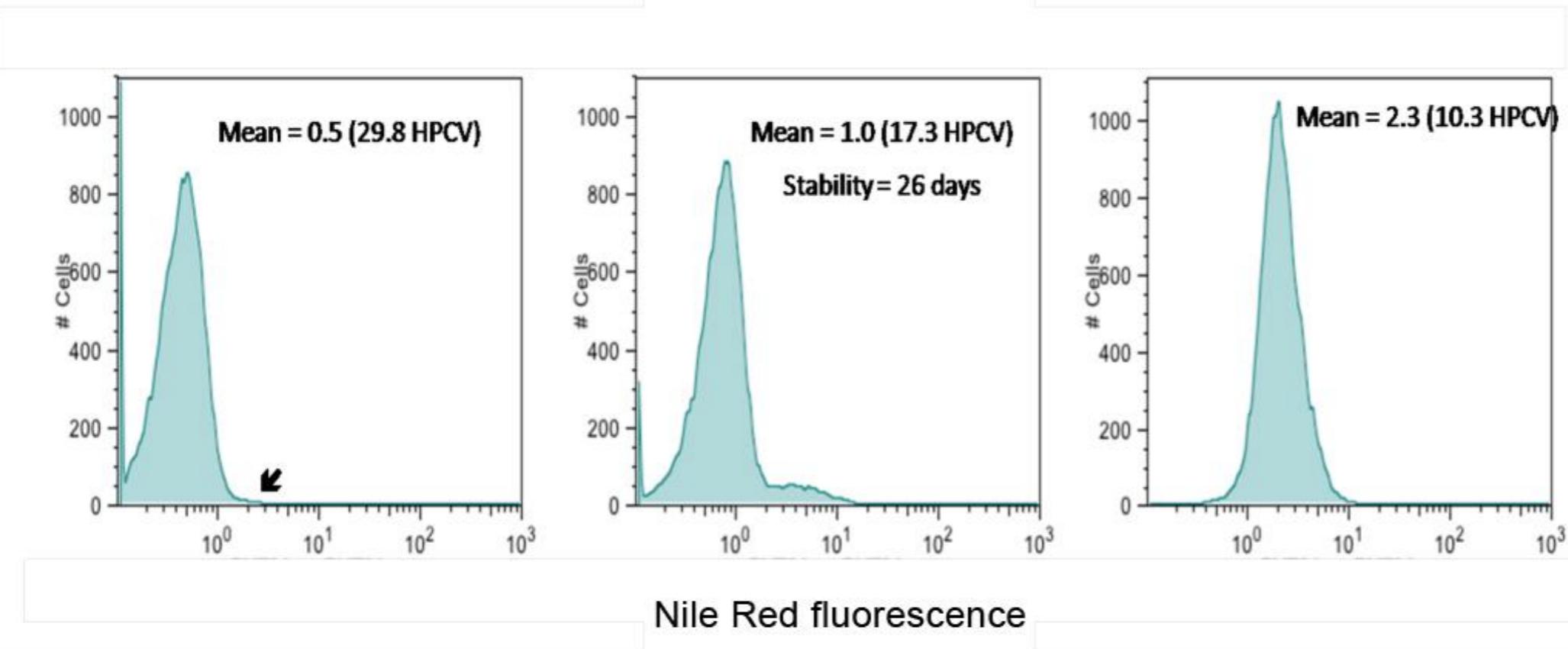
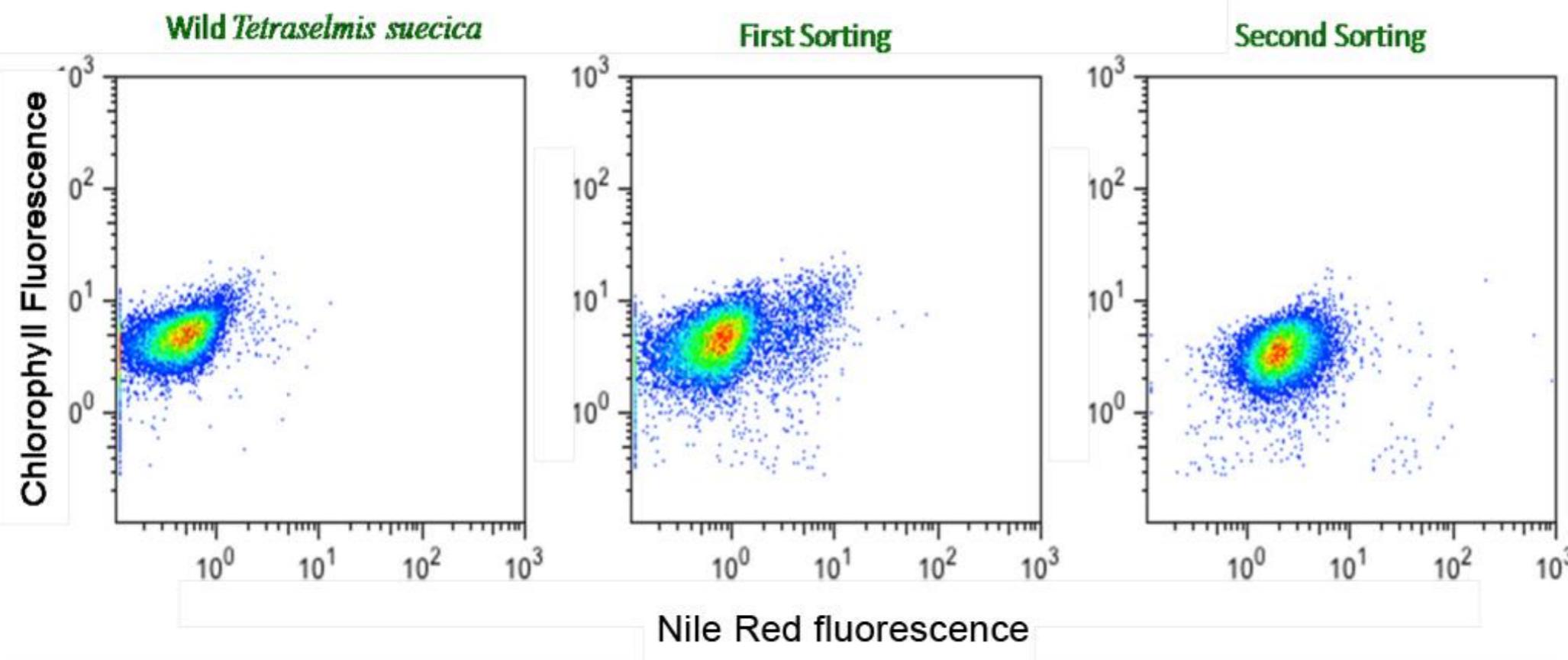


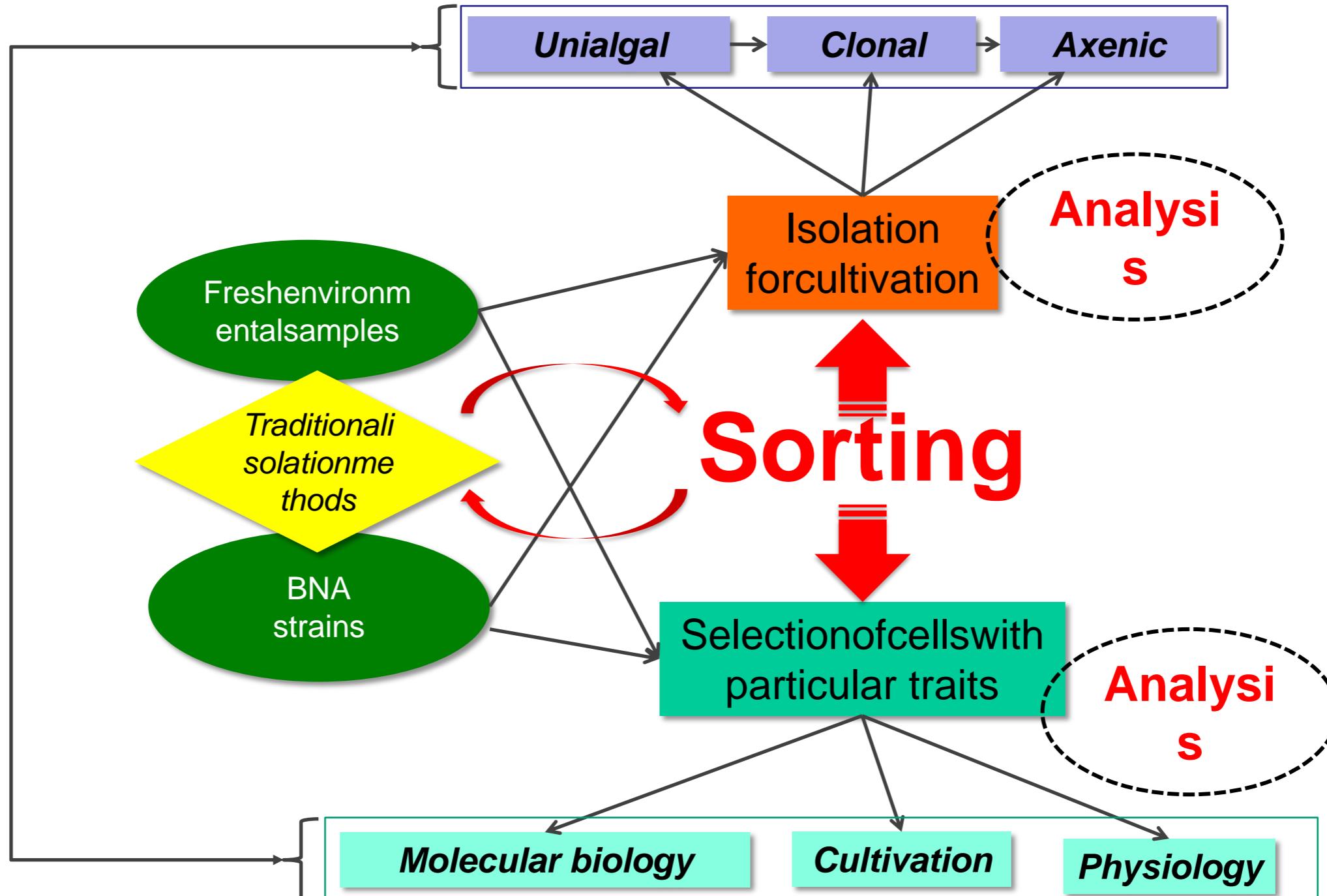


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Screening for hyperproductive strains - lipids

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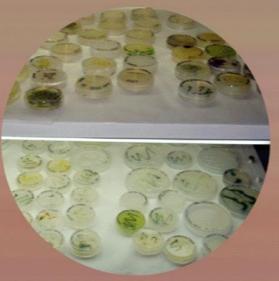




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NationalBankofAlgae - Marine BiotechnologyCenter

www.marinebiotechnology.org



Marine Biotechnology Center - BNA - ULPGC
Muelle de Taliarte, s/n - 35214 - Gran Canaria - Spain
Tlf: +34 928 133 290 - Fax: +34 928 132 830

www.marinebiotechnology.org



THANK YOU!