

PLEC DE PRESCRIPCIONS TÈCNIQUES

**CONTRACTACIÓ DEL SERVEI PEL DESENVOLUPAMENT D'UN
"MICROORGANISM CONCENTRATOR AND FLUORESCENCE READER PER
A L'INSTITUT DE CIÈNCIES FOTÒNIQUES MITJANÇANT PROCEDIMENT
OBERT SUBJECTE A REGULACIÓ HARMONITZADA**

NÚMERO D'EXPEDIENT: 2021.SE.001

CLAUSE 1. Description

Full-service contract for the development of a portable system including microorganism concentrator and fluorescence reader for microorganism detection in water.

CLAUSE 2. Overview

ICFO needs to develop a new microorganism monitoring system meeting bathing water testing requirements. Such monitoring system will use and be compatible with ICFO's proprietary microfluidics labelling system (MLS). ICFO has a preliminary laboratory version of the MLS, using fluorescence RNA probes based on Fluorescence In Situ Hybridization (FISH), which have been found as a reliable solution for the specific labelling of E.Coli and Enterococci which may give response to the water control authorities in terms of specificity. **ICFO thus subcontract an engineering company for the development of a portable system comprising two modules, a sample concentrator and a fluorescence reader, for microorganism monitoring in water.**

The concentrator module should be capable to process the large water volumes, in a high salinity range, from fresh, to brackish and sea water, increasing the representativeness of the analysis and enhancing the limit of detection of the reader by cell trapping filtration and elution of the raw sample. The fluorescence reader should be capable of measuring both labelled specific bacteria and phytoplankton (It should detect fluorescence emitted by microorganisms both naturally and by induced labelling techniques).

CLAUSE 3. Technical specifications

3.1 The fluorescence reader

The fluorescence reader module should be able to operate with at least two different fluorescence channels (wavelengths) for accurate counting of:

- 1) **phytoplankton** (below 50 micron size) through its auto-fluorescence with a **limit of detection equal or below 10 cell/mL, in a concentration operation range from 10 cell/mL to 10⁴ cell/mL.**
- 2) **specific labelled bacteria** using fluorescence RNA probes based on Fluorescence In Situ Hybridization (FISH), with a **limit of detection equal or below to 10 CFU/mL, in a concentration operation range from 10 CFU/mL to 10⁴ CFU/mL.**

In addition, the fluorescence reader should:

- 3) be able of reporting **real counts from individual fluorescence events**, or pulsed signals detected in the sensor, corresponding to individual target microorganism fluorescence emission being interrogated by blue or UV light source.
- 4) be **portable** with IP65 casing
- 5) be **compatible** with both the **MLS and the concentrator.**

3.2 The concentrator

The concentrator module should be able to perform an ad-hoc sample preparation to enhance reader's limit of detection by a concentration process,

- 1) achieving a **concentration factor from 10 to 10³**, using a filtration cell trapping media,
- 2) including **large volume sampling range (>1 liter)**.
- 3) The system is required to efficiently recover from the filter media in a suitable bacteria safe buffer. **Microorganism recovery efficiency in the elution process: > 50%** with gram negative bacteria and > 50% with gram positive bacteria.
- 4) Any associated **turbidity of this buffer may require a reduction from 10 to 100 NTU** (i.e. a preferred level of final turbidity to be below 5 NTU), **any solids separation applied should have no significant impact on cell trapping (bacteria recovery)** and the elution in small volume prepared in a suitable vial capable of transfer it to be labelled in the MLS or measured by the reader.
- 5) The concentrator should be **portable with IP65 casing**, self-contained within a housing suitable for being carried, fitted with all tubing with capability to be easily replaced, all pumping requirements for supply internal demands (buffer and other useful reagents) to perform the task, reagent reservoirs for use within the system, with programmable sample volume process and digital flow monitoring.
- 6) The concentrator needs to be **compatible with both the MLS and the reader**.

3.3 Combined module requirements

The required modules, concentrator and reader, in combination with the existing ICFO's MLS will:

- 1) process and analyse a **large water volume higher than 1 litre** for statistical relevance,
- 2) quantify live **E.Coli and Enterococci at minimum concentration levels of 1 CFU/100ml**,
- 3) quantify non-specifically live Phytoplankton, with a size less than 50um, at **minimum concentration levels of 10 cells/ml**.
- 4) possess a **time to result** for the concentration and reading quantification of the 3 target microorganism has to be no more than **1 hour**, in order to allow for a total of no more than 3h from sample to result (assuming the MLS takes a maximum of 2 hours to perform the labelling assay).
- 5) be compatible with different **Water matrices**: The concentrator and reader, in combination with ICFO's automated sample RNA labelling system, have to rapidly and **robustly** measure the concentration of specific bacteria and phytoplankton operating in a wide range of water matrices, from high salinity to low salinity, including **sea water, brackish water and fresh water with high matrix variability and very dependent on raining episodes**.

6) have **size and casing requirements of the modules**: The concentrator and the fluorescence reader modules should be integrated in separate industrial suitcases and externally connected to a PC laptop for system control and user interface. The dimensions should be around 40x30x20 cm for each module. **A compact and portable casing with proper IP65 sealing for dust isolation and against water bursts.**

7) be easily **mass produced**.

CLAUSE 4. Demonstrated know-how

Full-service contractor has to demonstrate know-how in

- 1) **Cytometry systems**
- 2) **Fluorescence labelling assays and biological sample preparation.**
- 3) **Water concentration methods including filtration and elution processes.**

CLAUSE 5. Scope of the service

The full-service contractor has to independently perform the following services:

1. System design.
2. System development.
3. Delivery of one demonstrator portable prototype of the system (two independent modules) suitable for field testing in the specified delivery time in clause 6, under mentoring of 1 year of validation.

CLAUSE 6. Starting and delivery time

The development of the systems will start the day after the signature of the contract resulting from the tender.

Completion time for both prototype modules **at the end of March 2022**.

CLAUSE 7. Target price

280.000,00 Euro (VAT excluded). Approximate breakdown:

Development costs	
Concentrator	110,000 €
Fluorescence Reader	170,000 €
TOTAL	280,000 €

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