Working session: Marine Biodiversity & Ecosystem Functioning
Flash talk

PhytoNumb3rs: An easy-to-use computer toolkit for counting microalgae by the Utermöhl method

ARPA PUGLIA

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Phytoplankton is recognized as a BQE and a biological descriptor in several European directives and national laws.


56/2008/EC
D. Lgs 190/2010

Descriptors
1 - Biodiversity
2 - NIS
4 - Food web
5 - Eutrophication/HABs blooms

The heterogeneity of data sources creates a barrier in terms of making connections within and among multiple domains of information.
The procedures used for phytoplankton analysis vary widely between research and monitoring groups, despite the numerous efforts to standardise phytoplankton data. The Utermöhl method (Lund et al., 1958) is the most widely adopted method to determine the abundance of phytoplankton assemblages.
PHYTONUMB3RS Tool Kit
A simplified visualisation of PhytoNumb3rs workflow

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PhytoNumb3rs: An easy-to-use computer toolkit for counting microalgae by the Utermöhl method

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Daniela Donadei, Marisa Florio, Emanuela Longo, Stefania D’Arpa, Flavia Maci, Sergio Ranieri,
Mariangela Spinelli, Annamaria Pastorelli, Nicola Ungaro

Regional Agency for the Environmental Protection and Promotion (ARAEN), Corso Tribunale 27, Rieti, Italy

Community meeting-Rome, 27-29 May 2019
<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Start of Analysis</th>
<th>Station Name</th>
<th>Analyzed By</th>
<th>Sedimentation Chamber V</th>
<th>Sedimentation Chamber N</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Sampling Date**

<table>
<thead>
<tr>
<th>Counting Field</th>
<th>Magnification</th>
<th>N° of Counting Fields/Transects/Whole Chamber</th>
<th>TAXA</th>
<th>Maximum Linear Dimension (N/M)</th>
<th>TOTAL</th>
<th>Number of Cells/Threads/Coenobia/Colonies</th>
</tr>
</thead>
<tbody>
<tr>
<td>Co-Counting Field</td>
<td>T=Transsect</td>
<td>F=Whole Chamber</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**TAXON LIST MARINE STRATEGY**

- Prorocentrum micans
- Prorocentrum gracile
- Prorocentrum lima
- Prorocentrum marinum
- Prorocentrum mexicanum
- Prorocentrum minimum
- Prorocentrum nanum
- Prorocentrum ovum
1. Upload SCDS

Insert the number of chamber

Detection limit:\n\[ \text{Detection limit} = -\ln(\alpha) \cdot \frac{\int \rho_{\text{true}} \, dV}{\int \rho_{\text{counted}} \, dV} \]
### Enumeration Data Sheet – OUTPUT Data Sheet

3. Copy for counting strategy

4. Create unified taxon list

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Unit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cell density</td>
<td>Cell/L</td>
</tr>
<tr>
<td>Number of cell counted</td>
<td></td>
</tr>
<tr>
<td>Lower confidence limit for taxon</td>
<td>Cell/L</td>
</tr>
<tr>
<td>Upper confidence limit for taxon</td>
<td>Cell/L</td>
</tr>
<tr>
<td>Quantitative Detection limit</td>
<td>Cell/L</td>
</tr>
<tr>
<td>Upper/Lower confidence limit (%) for total or groups</td>
<td>%</td>
</tr>
</tbody>
</table>

If a taxon has been counted below the detection limit the spreadsheet cell is highlighted in yellow.
Main advantages of PHYTONUMB3RS Toolkit

- Completely developed in Excel
- Automatically counts the number of cells belonging to the same taxon;
- Automatically counts the number of cells counted with the same counting strategy and the total number of cells counted in the sample;
- To Carry out quality control by entering the taxon name from a drop-down menu of user-defined taxon lists (reduction of typing errors and formatting errors);
- To Manage a new taxon entry (if the taxon name is not included in the list, the analyst can add it);
- To Calculate cell density uncertainty as for each taxa as for total (for quality assurance procedures);
- To Calculate the LOD;
- To Alert for taxa counted below the LOD
- To Export data in various templates (tables or lists)

**Phytonumb3rs** represents a first step towards harmonization of data and the promotion of standardized procedures for data management that will save time during database entry and storage. It makes possible to obtain high-quality databases, reducing random errors generated by the operator (typing, wrong names, etc.). The large-scale distribution of PhytoNumb3rs is advantageous because can improve the interoperability and integration of phytoplankton data collected by separate research and monitoring programs.
How can we improve PHYTONUMB3RS Toolkit?

Raw Data Sheet

- To Insert alert with advises for suitable counting strategies
- To Create a unified taxon lists of phytoplankton shared among different users

Enumeration Data Sheet – OUTPUT

- To Make the export data sheet function available to a wider range of users
- To Develop data processing workflow for re-using data for long term or spatial data analysis and creation of data reporting

Study cases in LifeWatch Italy VREs& MoBiLab – ARPA PUGLIA