EUROPEAN COOPERATION IN SCIENCE AND TECHNOLOGY

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Cyanobacterial blooms and toxins in water resources: Occurrence, impacts and management.

Short Term Scientific Mission (STSM)

Characterization and re-evaluation of selected Pseudanabaenaceae and Leptolyngbyaceae strains using polyphasic approach

Boris Aleksovski, M.S.

Institute of Biology,

Faculty of Natural sciences and Mathematics, Ss. Cyril and Methodius University , Skopje, Macedonia

Group leader: Prof. Svetislav Krstic

Objectives

The purpose of this STSM was characterization and re-evaluation of selected Pseudanabaenaceae and Leptolyngbyaceae strains using the modern polyphasic approach. In the present study we have characterized 9 new strains of *Pseudanabaena* isolated from Polar Regions, and we made re-evaluation of 15 selected members of Pseudanabaenaceae and Leptolyngbyaceae families present in culture collections, on the basis of combined molecular and cytomorphological markers. The results from the molecular evaluation of the selected strains were carefully compared with the performed morphological analyses, ultrastructure and the present information from ecology. The final goal was to examine whether the current system for classification of Pseudanabaenaceae and Leptolyngbyaceae match phylogenetic evidence and reflects evolutionary relationships.

Methodology

The complex work of morphological analyses, ultrastructure evaluation and use of molecular sequencing were the main methodological approaches in the study.

24 strains isolated from Polar Regions, or obtained from culture collections were carefully examined. The genomic DNA from the unialgal strains was isolated according to the modified protocol of Yilmaz et al. (2009). The 16S rRNA gene with the 16S-23S intergenetic segment as well as the rpoC1 gene were amplified and sequenced. The phylogenic trees were constructed by using the statistical method Maximum Likelihood, with Bootstrap support based on running 1000 replicates, using the Mega 6 software package.

Results

Our results showed not a perfect match between the morphological groups and the clusters from the phylogenic tree. However, in spite of the simplicity of these organisms, some morphological characters were present and recognizable for the phylogenic clusters, which supported the idea that these clusters are possible to be characterized morphologically, so the users of this system can utilize them for distinction.

Our results confirmed that the molecular evaluation of the selected strains more or less coincide with some morphological characters of the species, but unfortunately it seems that some other markers which are not that distinct and recognizable are more important for the taxonomic classification instead of the proposed ones. On the basis of our research we suggest that the presence of aerotopes, the morphology of the apical cell and the motility should be the most important morphological criteria for taxonomic classification of these groups. Contrary, as it seems, the criteria like cell shape, variability of cell length, width of the trichomes and presence of sheaths should not be considered as crucial markers for separation of the taxa in the Pseudanabaenaceae and Leptolyngbyaceae families.

Highlights

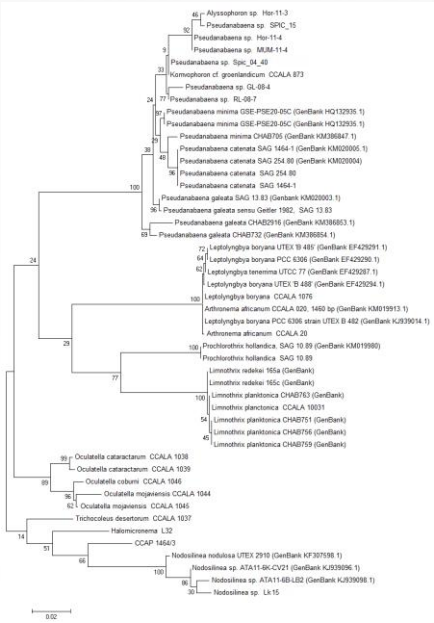
We find the results of our work quite progressive because of combining different criteria and because of the coincidences between some phenotypes and morphological groups with the clusters derived from molecular sequencing.

Host Organization

Institute of Botany , v. v. I. The Czech Academy of Sciences

Centre for Algology, Třeboň, Czech Republic

Group leader: Prof. RNDr. Jiří Komárek,



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Cyanobacterial blooms and toxins in water resources: Occurrence, impacts and management.

Short Term Scientific Mission (STSM)

Title: Methods in laboratory cultivation, isolation and characterization of cyanobacteria and their metabolites

Researcher

Jana PRIEBOJOVÁ, M.Sc.
Ph.D. student
Masaryk University
Faculty of Science,
RECETOX,
Czech Republic
priebojova@recetox.muni.cz

Klára HILSCHEROVÁ, Ph.D.
Assistant Professor
Ecotoxicology and risk assessment

Host Organization

Dr. Antonio QUESADA
Dpt. Biología
Universidad Autónoma
de Madrid
28049 Madrid
Spain

antonio.quesada@uam.es

Objectives

- Search for the retinoid metabolic pathways
- Search for genes involved in the synthesis of the enzymes responsible for carotenoid cleavage to retinoids
- Select cyanobacterial species for DNA extraction from UAM cultures collection
- Set up own PCR designed on the topic connected to the literature research
- Practise the DNA amplification

Methodology

- Cyanobacterial species (three from *Chroococcales* and seven from *Nostocales*) were cultivated in controlled laboratory conditions.
- For a DNA isolation were used UltraClean® Microbial DNA Isolation Kit (MO BIO Laboratories, Inc.).
- Thermocycler programs were created and Mastermix calculations were done based on found articles..
- The agarose gel was run at 90 V and after visualized in Gel Doc under UV light.

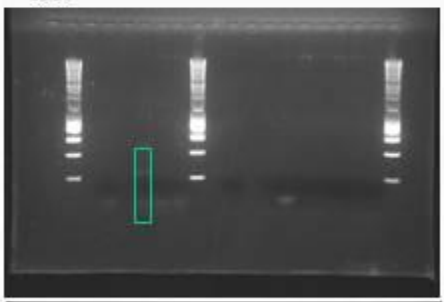
Results

- In selected species, we found just in case of *Microcystis aeruginosa* 2 bands in the gel.
- We repeated the PCR many times, but we didn't obtained the results that we got first time.
- We found the missing information about length of amplicons.

Highlights

The primers selected for this work, mastermix calculation and PCR conditions, were designed on these two artides.

Other info, pictures, graphs etc:



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