

Description of sequence files

Each file comprises all partial 16s rDNA sequences (mainly of cyanobacterial origin) of biological sand crust obtained from one sampling period, i.e. for the years 2010, 2011, 2012, in the fasta format. It can be directly used for alignments and can be opened in the word editor. The file name contains the station name, i.e. 69, 84, NS, and a hint of the relative location, i.e. ID for dune valley, SF for south-oriented dune slopes, and NF for north-oriented dune slopes.

Methodology

Study area and BSC sampling:

The three study sites (*Nizzana-South_NS*, *Nizzana-84* and *Nizzana-69*) are located along a precipitation gradient in the NW Negev, Israel. The southernmost study site *Nizzana-South* shows an average annual rainfall about 100 mm, followed by *Nizzana-84*, situated 12 km further north, with an average annual precipitation of about 130 mm and *Nizzana-69* (7 km further north) with an annual precipitation of about 170 mm. The annual rainfall is characterized by a high interannual variability and occurs largely between November and March.

Sampling for the characterization of cyanobacterial diversity was done March/April 2010, 2011 and 2012. At least three samples were taken from representative areas of the sampling plots. The top soil including the biological soil crust was extracted by pressing the basal part of an petri dish on the surface. Then, the petri-dish was closed and stored in the dark at 4°C. The biological soil crust from the entire sample was enriched by sieving, i.e. the loose sand was passed through a metal sieve (pore size 1 mm²) and crust pieces were collected. The isolated crust was homogenized under cooling using a mill (RETSCH, type MM400, Germany). The weight of the crust powder was then estimated (Kern, type ABT 120-5DM, Germany). Isolated crust pieces were then stored at -20°C before the analyses were performed.

DNA sequence analysis

Total DNA was isolated from 100 mg crust powder using the Invisorb Spin Plant Mini Kit (STRATEC Molecular GmbH, Germany). The DNA purification followed the instruction of the supplier. The final DNA was stored at -20°C. Appropriate dilutions of DNA were used for the PCR amplification internal fragments of the 16s rRNA gene with the PCR Master Mix (Quiagen GmbH, Germany). The cyanobacterium-specific primers (CYA 781 R: GACTACWGGGGTATCTAATCCCWTT; CYA 359 F: GGGGAATYTTCCGCAATGGG;

synthesized by Eurofins, Germany) described by Nübel et al. (1997) were used at an annealing temperature of 50°C. The reaction products were separated on 0.8% agarose gels and fragments of the expected size of approximately 400 bp were eluted from the gels using the NucleoSpin Gel & PCR clean-up kit (Macherey-Nagel, Germany) according to the manufacturer's instructions. The PCR fragments were then cloned into the plasmid pGEMT (Promega, USA). Recombinant clones of *E. coli* strain XL1 blue were selected from agar plates containing ampicillin, X-gal and IPTG. These clones were transferred in single wells of a Rotilabo Mikrotest Plate with U-profil (Roth GmbH, Germany) containing 200 µl LB-agar with 50 µg/ml ampicillin. The plates were sent to LGC-Genomics GmbH (Germany) which performed the micro titer plate sequencing using the following primers: M13-24F: CCAGGGTTTTCCAGTCACG and M13-24R: CGGATAACAATTTACACAGG. Per DNA approximately 40 independent clones were analyzed using forward and reverse sequencing primers.

All received sequences were inspected manually using the BioEdit Sequence Alignment Editor. Primer sequences were removed and sequencing errors corrected comparing the forward and reverse sequences.

Reference

Nübel U, Garcia-Pichel F, Muyzer G (1997) PCR primers to amplify 16S rRNA genes from cyanobacteria. *Appl Environ Microbiol* 63: 3327-3332