NOVEL COMBINED PHOSPHOLIPID AND 16S-rDNA PCR-SSCP ANALYSES TO CHARACTERIZE ENVIRONMENTAL BIOFILMS.

B. O. Ortega-Morales¹ and C. C. Tebbe².

¹Departamento de Microbiología Ambiental y Biotecnología, Universidad Autónoma de Campeche, Av. Agustín Melgar C.P.24030, Campeche, Camp., México. ²Institut für Agrarökologie, Bundesforschungstalt für Landwirtschaft, 38116 Braunschweig, Germany. E-mail: beortega@uacam.mx

Palabras clave: Tropical biofilms, molecular methods, biodeterioration

Introduction. Classical microbiological methods do not allow us to accurately assess diversity, biomass and other physiological attributes of microbial communities participating in biotechnological processes. Phospholipid fatty acids (PLFA) analysis is a community-level methodological approach that can provide useful information on the community composition, viable biomass and physiological status of microbial consortia. However, for microbial community analysis, nucleic acid-based methods have a better resolution than PLFA at the taxonomic level. PCR-single-strand-conformation polymorphism (SSCP) of partial genes encoding for RNA of the small subunit of ribosomes (SSU rDNA) is a genetic profiling technique that allows the fingerprinting of microbial communities. In addition, characteristic components (bands) of profiles can be selected and either directly sequenced or after cloning in Escherichia coli to identify the microbial members in a consortium.

A case study is provided to show the utility of this combined biphasic approach to characterize spatial patterns of fouling biofilm communities that colonize Mayan historic buildings.

Methodology. Biofilm samples were obtained from selected monuments. Phospholipid fatty acids (PLFA)¹ and PCR-single-strand-conformation polymorphism (SSCP) analyses were performed to fingerprint the extant microbial communities².

Results and discussion. A total of 35 sequences were detected, which fell within 5 bacterial phyla (Cyanobacteria, Proteobacteria, Bacteroidetes, Actinobacteria and Firmicutes), from which most of the sequences (68 %) belonged to Proteobacteria and Bacteroidetes. In general, similar bacterial groups (Cyanobacteria and Proteobacteria) and certain specific genera colonized most of the surfaces analyzed, although specific organisms appear to be particularly associated with interior habitats. Interestingly, most of the detected sequences and were related to halophilic bacteria, suggesting that substratum salinity may have selected for this type of metabolism on these Mayan monuments. The low level of similarity of our sequences as compared to that of extant sequences in public databases, suggests that much bacterial novelty, likely of halophilic nature, remains to be discovered³.

Conclusions. This study showed that environmental biofilms contain novel, not yet described microorganisms, whose biotechnological potential is currently unknown. In this case in terms of deteriogenic activity. The combination of PLFA analysis with SSCP is a novel powerful tool to gain insight attributed of microbial communities involved biotechnological process.

Fig. 1. Microbial community structure of epilithic biofilm using SSCP method on a polyacrilamide gel.

Acknowledgements. This work was supported by research grants CONACYT 33085-B and institutional support from UAC to B.O. O. M.

References