

It will also assess the presence of methanogenic Archaea and anaerobic dehalogenating bacteria in biofilms, developed inside anaerobic bioreactors used to remove chlorinated compounds from wastewater. In this project, traditional culture-based and microscopic techniques, chemical analyses, and molecular methods will be combined to better link microbial structure and function. Classical methods to characterize aerobic and anaerobic cells (fermentative products, microscopic exams, presence of anaerobic cofactors and enzymes) will be associated with molecular techniques, such as the use of fluorescent probes for hybridization with nucleic acids present in whole cells, in order to visualize single cells constituting specific populations in samples and the spatial position of various members in microbial communities. In addition, methanogenic cells and sulphate reducing bacteria will be detected by 16S rRNA probes and PCR amplification of 16S rDNA and DGGE. Molecular biology techniques will also be to determine the presence of specific catabolic genotype in the studied area.

This work was supported by the State of São Paulo Research Foundation (FAPESP) within the BIOTA/FAPESP The Biodiversity Virtual Institute Program (www.biotasp.org.br)

Pôster 188

TRANSPORT AND RETENTION OF COLIPHAGE T4 IN SOIL COLUMNS FROM A CEMETERY IN SÃO PAULO, BRAZIL Rodrigues, D.F.**; Matos, B.A.**; Gamba, R.; Pacheco, A.; Pellizari, V.P. Laboratório de Microbiologia Ambiental - ICBII. Universidade de São Paulo, USP

Abstract: Inadequate site and operation of cemeteries in urban areas may contaminate the water resources by microorganisms that grow during the process of corpse decomposition or by pathogenic organisms that may exist in the corpses. If the unconfined aquifer is contaminated in the internal area of the cemetery, that contamination may flow to surrounding areas and cause a health hazard to the people that use that water. Investigation studies at Cemetery Vila Nova Cachoeirinha, São Paulo, Brazil, have shown that the unconfined aquifer is contaminated by microorganisms. Columns under conditions of saturation and continuous flow were used to simulate virus migration in aquifers. Soil columns were prepared by carefully packing dry soil to the desired depth in 40 cm pyrex cylinder. Two columns were prepared, the first one was built up with high sand content samples and the other with high clay content samples. The coliphage T4 (ATCC: 11303-B4, host: *Escherichia coli* B) was used to simulate virus transport through the soil. The bacteriophage T4 was highly adsorbed to samples with high clay content, while adsorption to soils with high sand content was low. Biological (T4) and chemical (NaCl – 1%) tracers were injected into the column and the effluent was monitored for their presence. Relative concentration (C/Co) versus time curves of the effluent were plotted to study the retention of the coliphage by in the sand and clay columns. The high clay content columns retained more coliphage than the sand columns. Moreover, the coliphage took more time to be transported through the high clay content column. This study shows that coliphage T4 has been successfully used to simulate virus transport through soils.

Pôster 189

EVALUATION OF THREE QUANTITATIVE MICROTITER CYTOTOXICITY ASSAYS FOR *Escherichia coli* SHIGA TOXIN. Franzolin, M.R., Menezes, C.A., Lima, F.A., Trabuasi, L.R., Piazza, R.M.F. Laboratório Especial de Microbiologia – Instituto Butantan – E-mail: garoto@usp.br

Enterohemorrhagic *E. coli* (EHEC) has been the cause of many large diarrhea outbreaks all over the world. The disease caused by EHEC is called hemorrhagic colitis that is frequently associated with hemolytic uremic syndrome (HUS). Toxins produced by EHEC (Stx1 and Stx2) cause both bloody and hemolytic uremic syndrome, formerly designated Shiga-like toxins or verotoxins. Usually, the cytopathic effects of these toxins are detected by cytotoxicity assay in Vero cells, as a qualitative method. In order to compare qualitative and quantitative methods, three different stains were tested, neutral red, crystal violet and amido black. The cytotoxicity activity of *E. coli* was assayed by exposing several dilutions of culture supernatant to Vero cell monolayers in 96-microtiter culture plates. Stx1- or Stx2-positive *E. coli* O157:H7 were grown in two different culture media, centrifuged and the supernatant were filtered. Also, a French Press extract of Stx1-positive EHEC was tested. The cell monolayers were incubated for 72 hours with the samples and toxin potency was estimated by staining residual cells, followed by a visual