

# **Phylogenetic analyses of microbial populations from South African fly ash based on Denaturing Gradient Gel Electrophoresis (DGGE) profiles.**

**KA Reynolds<sup>1</sup>, AKJ Surridge<sup>2</sup>, TE Cloete<sup>2</sup> and D Cowan<sup>3</sup>**

<sup>1</sup>Eskom CR & D, Research and Strategy Division, Rosherville, Johannesburg, South Africa

<sup>2</sup>Department of Microbiology and Plant Pathology, University of Pretoria, Pretoria, South Africa

Fly ash is a waste product of the fossil fuel power industry. Crushed coal is subjected to stringent processing (temperature and pressure) resulting in the formation of highly alkaline fly ash. Thus, fresh fly ash has been considered to be a sterile product incapable of supporting microbial populations. In this study pro- and eukaryotic populations of fly ash were investigated using denaturing gradient gel electrophoresis (DGGE). Total DNA was extracted from fly ash samples of differing age. The 16S bacterial gene and a partial sequence of the eukaryotic internal transcribed spacer sequence (ITS2) of the rDNA operon were amplified by means of PCR. DGGE fingerprint profiles obtained were analysed before excising portions of the bands. Selected bands were then sequenced, analysed and compared with known sequences deposited in GenBank. Phylogenetic analyses were performed resulting in the grouping of these representatives according to the closest possible relative. Based on initial phylogenetic relationships it is clear that eukaryotes form the predominant population of fly ash. Preliminary identification of these representatives indicates that the microbial population in fly ash increases with time displaying differing predominant groups of organisms.

**Submitted for consideration in the World of Coal Ash 2007 Conference held May 7-10, 2007**