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# Method for extraction of high molecular weight quality metagenomic DNA from oil contaminated soil

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Hydrocarbons are problematic environmental pollutants due to bioaccumulations in both soil and water environments and slow biodegradation. Removal of these hydrocarbons is considered one of the main concerns with regards to bioremediation activities. Identification of microbial hydrocarbon genes is vital to bioremediation strategies. Efficient extraction of high molecular weight genomic DNA for downstream applications like next generation sequencing (NGS) and metagenomic library construction presents a major challenge. A common problem with DNA obtained from soil is the presence of humic and fulvic acids which co-extract with the DNA. Furthermore, traces of organic substances and heavy metals remain in the DNA extract leading to interference with molecular methods downstream. In order to study the diversity of a complex environment like a hydrocarbon-contaminated soil through culture independent techniques, efficient protocols that eliminate these contaminants and yield good quality DNA must be developed. This study describes a modified method for extracting genomic DNA from heavy oil contaminated soils. A general soil extraction method using CTAB was implemented however following the centrifugation of the soil and CTAB extraction buffer after incubation at a temperature of 65 °C, the supernatant was filtered through two layers of mesh guaze in order to remove the thick layer of oil floating at the top. Humic acid removal was also carried out using 2% CaCl2 on the crude DNA. The isolated crude DNA was separated on an agarose gel and gel electroelution was carried out on the excised gel fragments. Both the quality and quantity of isolated DNA was compared with existing methodologies and was found to be of high quality and an acceptable quantity. The maximum recovery of genomic DNA in the absence of substantial amount of impurities can be attributed to modifications in the isolation protocol and use of the gel electroelution technique.

## Biography

Cindy Baburam is a Lecturer and Researcher in the Biotechnology Department at the Vaal University of Technology and is currently pursuing her PhD, working under the supervision of N A Feto. Her Master's degree was conducted at the South African Sugarcane Research Institute in Durban, South Africa.

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