

TITLE: MOLECULAR CHARACTERIZATION OF BRAZILIAN ISOLATES OF *Mycobacterium vanbaalenii/austroafricanum* AND EVALUATION OF THE CAPACITY OF DEGRADING PYRENE.

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Polycyclic aromatic hydrocarbons (PAHs) are compounds commonly found in the environment as the result of the incomplete combustion of organic matter. PAHs are mainly produced as a byproduct on the industrial scale of exploration and refining of petroleum. Due to their complex structures and low solubility in water, they remain for long periods in the environment and are considered pollutants. The degradation of these compounds by microbial activity may occur via two paths: the dioxygenase (NID) and / or monooxygenases (CYP). The strain *Mycobacterium vanbaalenii*-PYR1 was described as the first microorganism capable of degrading pyrene, one of the high molecular weight HPAs. Sequencing of the genome of PYR1 strain revealed the presence of a specific spot denominated A region, which contains *nid* genes, related to the degradation of HPAs. In addition, another group of genes, responsible for the codification of monooxygenases (*cyp150*) were found to be dispersed in their genome and are also involved in the degradation of these compounds (initial step). In this work we investigated the presence of *nid* (*nidA*, *nidB*, *nidA3* and *nidB3*) and *cyp* (*cyp150*) genes by PCR in five isolates of *Mycobacterium vanbaalenii/austroafricanum* isolated from aquatic environments and also evaluated the degradation of pyrene using the double-layer plates phenotypic test. In addition, the clonality of the isolates was verified by Pulsed Field Gel Electrophoresis (PFGE) with *DraI* and *XbaI* enzymes. The PFGE results revealed that two of five isolates analyzed presented indistinguishable profiles, suggesting clonal origin. The presence of the *cyp150* gene was evidenced in the DNA of all the isolates. In contrary, none of them generated amplification products of the genes encoding the analyzed dioxygenases. Four of five isolates presented a clear zone of pyrene degradation on the double layer test. In conclusion, the majority of isolates from this study had the ability to degrade pyrene which may be an indication that the A region of these isolates may be distinct from the A region present in the genome of the reference PYR1 strain.

Keywords: *Mycobacterium*, dioxygenases, monooxygenases, degradation, pyrene.

DEVELOPMENT AGENCY: CAPES