

TLO-7. Molecular cloning and functional characterization of a catalase gene from a *Bacillus safensis* strain isolated from an oil-impacted brazilian mangrove sediment

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Introduction. Mangroves are rich environments which harbor a wide diversity of life. They generally constitute an extreme habitat for microorganisms due to their high salinity, anoxic conditions, lack of nutrients and pH variations. Beyond these harsh characteristics, the bertioga city mangrove (brazil) was affected by an oil spill in 1983, which promoted a selection of microorganisms adapted to such environment. Adapted microbial communities may constitute important source of active molecules, however, researches involving microbial diversity in these environments are scarce, making the knowledge of new microorganisms in mangroves an urgent issue. The aim of this work was to characterize a novel catalase gene from *Bacillus safensis* strain ccma-560 isolated from bertioga mangrove.

Materials and methods. The catalase gene was discovered by *in silico* analysis of img/m annotation of its genome, followed by gene PCR amplification using specific primers, and subsequent cloning, expression and purification. The catalase gene was cloned into pet28a expression vector which contains a 6x-his-tag and transformed into *E. coli* bl21(de3) rosetta cells. The recombinant enzyme was overexpressed in lb broth supplemented with 30 µg/mL of kanamycin and 34 µg/mL of chloramphenicol for 16 h incubation at 25°C/200 rpm. Induction of protein expression was achieved by addition of 0.4 mM iptg and purification was performed by affinity chromatography. Catalase activity was accessed using hydrogen peroxide as substrate.

Results. The overexpressed enzyme was active in a broad pH (6-10) and temperature (22°C-60°C) range.

Conclusions. Results indicated alkali and thermotolerant behavior of catalase. Further functional and structural characterization may provide valuable data about the versatility of this new enzyme from an extremotolerant microorganism isolated from mangrove sediments. Acknowledgements: fundação de amparo à pesquisa do estado de são paulo -fapesp. Processes: 2010/51981-3, 2011/50809-5, 2013/16813, 2011/50268-4, 2011/10378-5.