

Cloning and purification of a lipase from *Geobacillus thermoleovorans*

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Geobacillus thermoleovorans is a bacterium isolated from 3.1 km below the surface in the East-Driefontein mine and was shown to have exhibit lipase activity when induced with olive oil, tributyrin and stearic acid under growth conditions. The gene coding for the signal peptide and mature lipase was amplified and cloned into pGEMT-easy and showed high similarity to lipase from other *Geobacillus* species when it was sequenced. The gene was modified to include the mature lipase with either a C-terminal or N-terminal histidine tag. The N-terminal histidine tag could be removed using the thrombin cleavage site incorporated into the gene, yielding the detagged lipase. The characteristics of the clones correlated with that of the native lipase isolated from the host with optimum pH 8-9 and optimum temperature of 65C. Using monolayer techniques it was shown that the N-tagged and detagged lipases had a stereo- and regio preference for the *sn* 1 and 3 positions. It was interesting to note that the N-terminal histidine tag enhanced the activity of the lipase when tested against 1,3 dicaprylyn at a constant surface pressure of 15 mN.