

## Cloning, expression and characterization of a novel cold-adapted GDSL family esterase from *Photobacterium* sp. strain J15

### Abstract

The gene encoding for a novel cold-adapted enzyme from family II of bacterial classification (GDSL family) was cloned from the genomic DNA of *Photobacterium* sp. strain J15 in an *Escherichia coli* system, yielding a recombinant 36 kDa J15 GDSL esterase which was purified in two steps with a final yield and purification of 38.6 and 15.3 respectively. Characterization of the biochemical properties showed the J15 GDSL esterase had maximum activity at 20 °C and pH 8.0, was stable at 10 °C for 3 h and retained 50 % of its activity after a 6 h incubation at 10 °C. The enzyme was activated by Tween-20, -60 and Triton-X100 and inhibited by 1 mM Sodium dodecyl sulphate (SDS), while  $\beta$ -mercaptoethanol and Dithiothreitol (DTT) enhanced activity by 4.3 and 5.4 fold respectively. These results showed the J15 GDSL esterase was a novel cold-adapted enzyme from family II of lipolytic enzymes. A structural model constructed using autotransporter EstA from *Pseudomonas aeruginosa* as a template revealed the presence of a typical catalytic triad consisting of a serine, aspartate, and histidine which was verified with site directed mutagenesis on active serine.

**Keyword:** Cold-adapted; GDSL family; Esterase; Cloning; Characterization; *Photobacterium*