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A point mutation at the vicinity of active site of a lipase enhances the protein thermostability of a mutant several fold

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In the present investigation, a thermostable mutant lipase was generated employing directed evolution approach. The mutant exhibited 144 fold enhanced thermostability over the wild type enzyme at 60°C. The mutant enzyme showed high catalytic efficiency (kcat/Km). Biophysical investigation employing circular dichroism spectroscopy demonstrated that the mutant enzyme retained its secondary structure up to 70–80°C, whereas the Wt type protein structure was completely distorted above 35°C. In addition to this, the intrinsic tryptophan fluorescence conducted for the Wt and mutant enzyme also displayed variation in conformational changes during temperature dependent unfolding. By comparing the modelled structure of the Wt and mutant enzyme we found that the mutant N355K mutation exhibited extensive H-bonding (Lys355 HZ1\OE2 Glu284) with a distance 2.44 Å, whereas the Wt enzyme has not shown such H bonding interaction.

Biography

Pushpender K. Sharma, presently working as Assistant Professor at Sri Guru Granth Sahib World University, Fatehgarah sahib, Punjab, India. He completed is Postdoctoral fellowship in department of biotechnology at IISER, Mohali, India.

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