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Optimization of alkaline protease production by *Bacillus cereus* FT 1 isolated from soil

B. Asha[1,2], M. Palaniswamy[1*]

[1]Karpagam University, Karpagam Academy of Higher Education, Coimbatore, Tamilnadu, India.
[2]SIAS Centre for Scientific Research, Vazhayoor, Malappuram, Kerala, India.

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ABSTRACT

Alkaline proteases have high commercial value and find multiple applications in various industrial sectors. The present study intended to isolate a suitable bacterium for alkaline protease production. Protease producing bacteria were isolated from organic waste containing soil, screened for protease production on skim milk agar plates and confirmed the protease production through protease assay. The bacterial isolate showing highest alkaline protease production was selected and identified by microscopic, macroscopic, biochemical and 16 S RNA phylogenetic analyses as *Bacillus cereus* FT 1. Maximum enzyme production by the isolate was obtained at 35°C; pH, 9.5; 2% lactose as a carbon source and 3.5% casein as a nitrogen source after 48 h of incubation. Among the various surfactants tested, tween 20, tween 80 and poly ethylene glycol were found to be increasing the protease production by the isolate. Mn^{2+}, among the metal ions tested tremendously increased the protease production. The best organic solvent for protease production was found to be petrol. With all the optimised cultural conditions, maximum enzyme activity was found to be 187 U/mL and the enzyme was a promising one for detergent industry as an additive enzyme.

INTRODUCTION

Proteases are enzymes with many physiological roles in all living organisms including cell growth and differentiation (Barrett *et al.*, 2001; Burhan *et al.*, 2003). They are also recognized as industrially important and occupy almost 60% of the total enzyme market (Gupta *et al.*, 2005; Chu, 2007; Verma *et al.*, 2011). Among the proteases, alkaline proteases have the applications in industries like laundry detergents, pharmaceutical, leather, food processing and proteinaceous waste bioremediation (Bayoudh *et al.*, 2000). They are highly active and stable under alkaline conditions (Maurer, 2004; Saeki *et al.*, 2007).

Proteases produced by bacteria are most significant as their properties can be easily modified through genetic manipulations to suit their various applications (Najafi *et al.*, 2005). Most of the commercially important alkaline proteases are produced by *Bacillus* spp. The alkaline proteases derived from *Bacillus* sp. are highly active and stable at different pH and temperature ranges, broad substrate specific, and can be easily purified with low cost (Maurer, 2004; Haddar *et al.*, 2009; Jellouli *et al.*, 2009). Detergent industry requires efficient, environmental friendly and economical strategies for unwanted protein degradation. Alkaline proteases in detergent formulations can act against proteinaceous stains like blood, food and grass stains (Hameed *et al.*, 1996; Smulders *et al.*, 2002; Huang *et al.*, 2003; Wang *et al.*, 2007; Kalpana Devi *et al.*, 2008).

The optimization of different fermentation parameters like nitrogen and carbon source, media pH, incubation temperature, agitation and incubation time can enhance the yield of industrially useful enzymes (Huang *et al.*, 2003; Tobe *et al.*, 2005; Boominadhan *et al.*, 2009; Aruna *et al.*, 2014; Lakshmi *et al.*, 2014). Although, industrially applicable protease enzymes have been identified from different sources, most of them could not resist drastic environmental changes and most of the sources are incapable to produce required quantities to fulfil industrial demands. So, new bacterial strains that can withstand harsh environmental conditions should be isolated for the enhanced production of such enzymes. The present study is aimed at the

*Corresponding Author
M. Palaniswamy, Dean, Faculty of Arts, Science and Humanities, Karpagam University, Karpagam Academy of Higher Education, Coimbatore, Tamil Nnadu, India. E-mail: m.palaniswamy @ gmail.com