CHAPTER I

INTRODUCTION

Lignocellulose,the most abundant and renewable organic compounds in nature, comprises of average 40% cellulose, 33% hemicellulose and 23% lignin by dry weight (Sa-Pereira *et al.*, 2002). The cellulose is composed of D-glucose units linked together to form linear chain via β-1,4-glycosidic linkages (Salmon, 1997). Cellulolytic enzymes, cellulases, are a group of enzymes which hydrolyse β-1,4-glycosidic linkages in cellulose. The cellulolytic enzymes is composed of at least three different enzymes. Endoglucanases (1,4-β-D-glucan -4-glucanohydrolase, E.C. 3.2.1.4) attack randomly internal linkages within the cellulose chain, creating free chain ends. Exoglucanases (1,4-β-D-glucan cellobiohydrolase, E.C. 3.2.1.91) hydrolyse cellulose from the free chain ends creating mainly cellobiose as an end product. β-Glucosidases (E.C. 3.2.1.21) hydrolyze the small oligomer including cellobiose to produce glucose. The cellulolytic enzymes act together in synergism. (Beguin and Aubert, 1994; Harjunpaa, 1998).

Complete set of the enzymes are efficiently degrade native cellulose. White-rot and soft-rot fungi produce the most efficient cellulase systems (Clarke et al., 1997). Extracellular cellulases from several bacteria have been studied and characterized *e.g. Clostridium thermocellum, Caldocellum saccharolyticum* and *Acidothermus cellulolyticus* (Bergquist *et al.*, 1999), *Bacillus subtilis* (Chan and Au, 1987), *Cellulomonas cellulans*, *C. fimi* (Lednicka *et al.*, 2000), *Ruminococcus flavefaciens* (Aurilia *et al.*, 2000), *R. albus* (Ohara *et al.*, 2000), *Acetivibrio cellulolyticus* (Ding *et al.*, 1999) and *Acetivibrio cellulosolvens* (Khan *et al.*, 1984). The best studied bacterial cellulases are *Cellulomonas fimi* and *Thermonopora fusca* cellulases (Irwin *et* al., 1998).

Due to increasing demands for more environmental friendly methods, the use of enzymes instead of polluting chemicals has significantly increased during the past two decades (Harjunpaa, 1998 ; Guedon *et al.*, 2001 ; Patel *et al.*, 2006). Cellulases have been used in several industries, for example, pulp and paper industry (Muzariri *et al.*, 2001) enzymatic pretreatment of pulp improves the mechanical properties of the wood fiber, leading eventually to better paper quality. Cellulases have been extensively utilized for extraction of valuable components from plant cells, improvement of nutritional values of animal feed, and preparation of plant protoplasts in genetic research. However, such applications require cellulase (s) with particular properties, *e.g.* active under high temperature and/or alkaline condition. Most of industry processes are carried out at high temperature, so that thermostable enzymes would give an advantage. Bacterial cellulases are generally higher thermostable than fungal cellulases. Most cellulases from fungi have pH optima between 4 and 6 (Zhu *et al.*, 1982 ; Yazdi *et al.*, 1990), while bacterial cellulases active at alkaline pH have been reported (Ruttersmith and Daniel, 1993). Nan province is located in northern part of Thailand. Seventy-five percent of the area is covered by enriched forests where several important rivers of the country are originated. Therefore, soil samples in Nan province is interesting and challenging resources for a discovery of novel cellulase-producing bacteria.

The main objectives of this present study are as followed:

1. To isolate and screen cellulase-producing bacteria from soils in Nan province, Thailand.

2. To identify and characterize the cellulase-producing bacteria from soils based on their phenotypic and chemotaxonomic characteristics including 16S rDNA sequencing.

3. To characterize cellulase production of the selected isolates based on the effects of pH and temperature.