CHAPTER I

INTRODUCTION

Apis mellifera Linnaeus, 1758 means a honey-bearing bee. It is indigenous only in Europe and Africa. Later, this species has been introduced from Europe into almost every country in the world (Ruttner, 1988). *A. mellifera* is highly eusocial insect and is divided into three levels in each colony. It is a valuably economic insect and can be managed in hive (Crane, 1999). This brings to a convenience to collect interesting samples. In 1997, Gregorc and Bowen studied phosphatase enzyme, especially in acid phosphatase (AC) and were able to detect cell death in midgut of *A. mellifera* larva. It may be possible that the activity of alkaline phosphatase (AP) is the same as the activity of AC but in basic condition. AP is found in most of species from bacteria to mammals. Most of AP is located in periplasmic space of membrane in prokaryotic cells but is found in glycoprotein in membrane or in lysosome of eukaryotic cells (McComb *et al.*, 1979). In human, four isoenzymes discovered in placental, intestinal, placental-like germ cell, and non-specific form at liver/bone/kidney have been recorded. AP is important to determine disorder condition in several organs of human or mammal (Henthorn *et al.*, 1987; Weiss *et al.*, 1986; Brain *et al.*, 1988). In insect, *Musca domestica*, the AP activity is found to be the highest in young larva stage. After that, the AP activity will decrease and become stable in adult stage (Barker and Alexander, 1958). The AP activity in *Drosophila melanogaster* plays roles in forming cuticle and is the highest in larva stage (Schneiderman *et al.*, 1966). The AP activity is always found in brain of several insects such as in *Periplaneta amercana* (Verhaert *et al.*, 1990), in *D. melanogarter* (Yang *et al.*, 2000). Furthermore, the AP activity is found at epithelial cells of appendages of *Schistocera americana* (Chang *et al.*, 1993) and at epithelial cell in midgut of *Bombyx mori* larva (Itoh *et al.*, 1991).

The objective of this research is to study the AP activity in workers of *A. mellifera* at various developmental stages. The research is divided into three parts. First, the activity localization in tissue in each developmental stage of *A. mellifera* was studied. The AP activity may be found in all types of tissue, especially at cell membrane. Second, a partial DNA sequence of *AP* and comparison homology of *AP* in *A. mellifera* with other organisms was determined. The partial DNA sequence was amplified by polymerase chain reaction (PCR) with AP1 and AP2 primers designed from conserved regions of amino acid of AP in several organisms. It may be mostly similar to *D. melanogaster*. Last, the AP activity was assayed by hydrolysis reaction of crude extract and polyacrylamide gel electrophoresis (PAGE). Pattern of the AP activity may be the highest at larva developmental stages.

In histochemical study, techniques may be applied to study AP in other honeybees. The primers may be used as in RT-PCR in order to confirm a pattern of *AP* expression in *A. mellifera* and other honeybees. In future, the AP of *A. mellifera* will be purified to determine characterization and specificity.