CHAPTER I
INTRODUCTION

Problem Statement

Thiopurine S-methyltransferase (TPMT, EC 2.1.1.67) is a cytoplasmic enzyme that catalyzes the S-methylation of aromatic and heterocyclic sulfhydryl compounds such as anticancer agents, 6- mercaptopurine and thioguanine, and the immunosuppressant azathiopurine. 6-Mercaptopurine (6-MP) is a prodrug with no intrinsic anticancer activity, and required intracellular conversion to thioguanine nucleotides (TGNs) which subsequently incorporated into DNA, producing antiproliferative effects. In standard treatment protocols for children with acute lymphoblastic leukemia (ALL), 6-MP is administered as a daily oral dose for a majority of 2-3 years of maintenance therapy. Myelosuppression is a major toxicity. For 6-MP, TGNs are formed by a multi-step pathway which is initiated by hypoxanthine phosphoribosyl transferase (HPRT). Alternatively, 6-MP is also methylated by thiopurine methyltransferase (TPMT) to methylmerpactopurine (MMP) or oxidised to thiouric acid by xanthine oxidase (XO). Metabolism via either TPMT or XO decreases the formation of active TGNs (Mcleod et al., 2000).

Levels of TPMT activity are importantly controlled by a common genetic responsible for inter-individual differences in thiopurine toxicity and therapeutic efficacy. Study in healthy Caucasian population shows a trimodal distribution of TPMT activity , 89-94% having a high activity with a loss of efficacy, 6-11% having an intermediate activity, and 1 in 300 having a low activity which develops severe myelosuppresion when treated with "standard dose" of 6-MP. The doses of 6-MP need to be significantly reduced by 8-15 folds in patients with undetectable TPMT activity. Several mutant alleles responsible for TPMT deficiency have been described and eight TPMT alleles have been identified. Three alleles, which are *TPMT*2, TPMT*3A* and *TPMT*3C*, appear 80-95% in intermediate or low enzyme activity cases (Weinshilboum, 2001).

The mutant alleles of TPMT have inter-ethnic variability with different frequency and pattern among various ethnic populations. For example, Southwest Asians (Indian,

Pakistani) have a low frequency of mutant TPMT alleles and all of the mutant alleles identified to date as *TPMT*3A*. Frequency of mutant alleles in East and West African populations are similar to the Caucasians. All mutant alleles in the African populations are *TPMT*3C* while the African-Americans have three-types of *TPMT*3C* (2.4%), *TPMT*2* (0.4%) and *TPMT*3A* (0.8%). Mutant alleles in South-East Asian population is identified as *TPMT*3C* (Mcleod et al., 2000). The study of TPMT activity frequency distribution in Thailand have two reports. First, Parida (1996) found that TPMT activity showed trimodal distribution, high (3.72%), intermediate (95.35%), and low (0.93%). The other, Suradej (2000) shown that genetic variation of TPMT activity exhibited bimodal distribution, high (89%) and intermediate (11%).

However, the information of TPMT genetic polymorphism in Thailand is scarce and no correlation. This study aimed to determine the frequency distribution of TPMT enzyme activity together with variation of mutant alleles studied in acute lymphoblastic leukemia children.

Hypothesis

Frequency distribution activity of TPMT is correlated with variation of mutant alleles found in ALL Thai children.

Objective

1. To determine the frequency distribution of TPMT activity in ALL children.
2. To study the variation of mutant alleles controlling TPMT activity ALL children.

Expected outcome

1. Valid information on the frequency distribution and variation of TPMT mutant alleles in Thai ALL children.
2. Application of information in (1) for adjusting 6-MP dose to maximize the efficacy and minimize the toxicity.