I. INTRODUCTION

Leprosy is a chronic infectious disease of human caused by intracellular bacteria, *Mycobacterium leprae*. The clinical manifestations of leprosy are associated with the loss of sensory and motor function of affected nerve, leading to anaesthesia, contractures, loss of pain reflex, and ultimately to ulceration and gross deformities, particularly of hands and feet (Hasting, 1985). Most individuals infected with *M. leprae* do not develop disease, whereas a few of infected persons develop various forms of leprosy. A number of different forms of leprosy can be distinguished into five classes according to clinical and histologic features (Ridley and Jopling, 1966). The first pole of leprosy is tuberculoid form (TT). This form is characterized by isolated lesions with few bacilli, low specific antibody titers and strong specific cellular immune responses. The second pole is lepromatous leprosy (LL). Lepromatous leprosy patients have many lesions spread throughout the body and numerous *M. leprae* can be found in skin lesions. This pole does not exhibit specific cell-mediated immune response but displays good humoral response that is not protective. Between these two poles, the three borderline classes: borderline-tuberculoid leprosy (BT), borderline leprosy (BB) and borderline-lepromatous leprosy (BL), show variable manifestations in between of TT and LL forms of leprosy.

It is believed that *Mycobacterium leprae* itself is virtually non toxic. The differences of clinical manifestations in patients that are exposed to the same infective agent depends largely upon host immune responses directed against the antigenic substance liberated from bacterial cells rather than directly due to *M. leprae* itself (Godal, 1974). Variation in the development of various clinical forms of leprosy is thought to be due to host factors, especially host immune response. For a long time, HLA and HLA-linked genes were considered to be candidates for susceptibility genes or to contribute to the development of various forms of leprosy. In earlier studies of HLA class I (HLA-A and HLA-B) and leprosy in various populations, it was found that different HLA class I antigens were associated with leprosy or types of leprosy (Dasgupta et al., 1975; Takata et al., 1978 and Rea et al., 1976). In Thailand, Youngchaiyud et al. (1977) found significantly increased frequency of HLA-Bw40 antigen in TT and LL patients as compared to controls,

whereas Greiner et al. (1978) found that HLA-B7 antigen was significantly increased in LL patients in northern Thailand.

Later, HLA class II genes were considered to contribute to the varied clinical manifestation of leprosy. This is because HLA class II molecules are known to play a central regulatory role in the immune response and they are mainly expressed on cells of immune system and function in the presentation of antigens to helper T cells. As a consequence of antigenic stimulation, helper T cells produce various cytokines that are necessary for cytotoxic T cells, B lymphocytes and macrophages to kill their target cells, produce antibodies, and kill intracellular parasite, respectively. Thus, T helper cells and HLA class II molecules have an indispensable role in the generation of immune response. With this consideration, De Vries et al. (1988) proposed the immune response gene hypothesis to explain differences in the susceptibility and manifestations of leprosy. It was thought that different antigenic epitopes of *M. leprae* are presented by the products of different HLA class II alleles to functionally different T cell subsets, resulting in either protective immunity or immunopathology. Many studies performed in several ethnic groups found that HLA-DR2 or -DR3 antigens were associated with clinical manifestation of tuberculoid leprosy, while HLA-DQw1 antigen was associated with lepromatous leprosy (De Vries, 1980; Miyanaka et al., 1981; Ottenhoff and De Vries, 1987; Gorodezky et al., 1987; Ottenhoff et al., 1984). In a sole study done in Thailand, HLA-DR2 and HLA-DQw1 antigens was strongly associated with the tuberculoid form of leprosy (Schauf et al., 1985).

In these studies, the serologic method that could distinguish only three and twelve allelic forms of the HLA-DQ and HLA-DR locus, respectively, was employed. Recent advances in molecular genetic methods have shown that the number of HLA class II alleles is greater than those detected by serological methods. Several methods can be used to analyze the polymorphism of HLA genes, such as DNA sequence analysis, restriction fragment length polymorphism (RFLP) and sequence specific hybridization. DNA sequencing is the most accurate method for the characterization and identification of polymorphic class II alleles, but, because of technical complexity, it is not suitable for large-scale population studies. However, the data from sequence analysis have been employed to set up more efficient and rapid methods for distinguishing multiple alleles of HLA class II loci. For example, the RFLP analysis can be used to detect different HLA class II alleles

but is limited to only some specificities (Bidwell, 1988). Most recently, the DNA typing procedure employing polymerase chain reaction and sequence-specific oligonucleotide (PCR-SSO) hybridization was developed. This technique is relatively simple and suitable for large-scale population studies; it can distinguish upto nine and seventeen alleles, respectively, of the HLA-DQA1 and -DQB1 loci.

In order to provide further support for the hypothesis that HLA class II genes are involved in clinical manifestation of leprosy, we used the PCR-SSO hybridization to determine the exact allele of HLA-DQ genes that may possibly be associated with tuberculoid and lepromatous forms of leprosy in northern Thailand.