Chapter 1 Introduction



Like the other tropical countries, forest degradation and fragmentation have been progressive in Thailand. Consequently, most wildlife habitats were degraded or eliminated. The surviving population which is been small and fragmented potentially losses the future ability to survive and evolve. Their conservation will require far more management than was necessary in the past. To develop an effective conservation management, various biological disciplines is necessary. Unfortunately, the Thai biota is still rather poorly known, making it difficult to set conservation priorities (Woodruff, 1991).

The Red Junglefowl, *Gallus gallus*, an important bird in evolution of human civilization and the main genetic stock for the world's lineage of domestic chicken (Fumihito et al., 1996), has stood on this situation. The Red junglefowl is one of four species of the world's Junglefowl naturally distributed in only the warmer part of Asia. Two out of five subspecies of this species which are distinguishable by their earlobe color and their distribution, partially inhabit in Thailand. *G. gallus gallus* with whitish ear spot has been found in the eastern part of the country whereas other red earlobe, *G. gallus spadiceus* inhabits in the northern forest from Lao border down to southwest along the Myanmar border through the Malay peninsula. Habitat degradation and fragmentation, hunting as well as geographical difference in habitat between Indo-China continental and Malay peninsula may be the causes of random genetic drift and genetic differentiation between local populations of the red ear lobe subspecies in northern and southern habitat.

The understanding of population genetic is necessary for evolution biology, breeding management and conservation of interested species. When combined with demographic, ecological, behavioral, and physiological characteristics of interested species, genetic data has emerged as a unifying component for interpreting past history, present status, and future prospects of threatened population. To study the population genetics, molecular method has replaced the conventional method which concentrate on easily detected variations including morphometric characters, blood groups or blood proteins and chromosomal inversion. Although these variations are important variation but they do not allow the estimation of total genetic variation of natural population. On the contrary, molecular techniques involve the examination of the direct translated products or the genes themselves so that the total genetic variation is clarified. In addition, interpretion of molecular data is relatively simple and well understood.

Unfortunately, previuos studies of the Red Junglefowl had been concentrated on taxonomy, morphology, ecology, behavior and its relationship among domestic chicken. Although, molecular techniques are now available to address the genetic questions in various species but molecular studies of this representative species of order Galliformes are relatively scarce. There are few researches using molecular techniques to examine the relationship between domestic fowl and the Red Junglefowl (Fumihito et al, 1994; 1996; Okada, 1994). Recently, Boripat Siriaroonrat (1997) used the nucleotide sequence of the mitochondrial control region to investigate genetic variation of these two subspecies of Thailand. Phylogenetic analysis shown a different branches between two subspecies whereas sequence divergence within each subspecies seem to be low. On the other hand, molecular markers and genetic data of chicken, a domestic descendant of this species, are well studied.

Hence, in order to investigate genetic variation among individuals within and between local populations of the red ear lobe subspecies exist in northern and southern habitat, chicken microsatellite markers have been used for two reasons. First, there are a number of available microsatellite markers of commercial domestic fowl, a closely related descendant of the Red Junglefowl. Using these markers to investigate genetic diversity of natural populations of the Red Junglefowl is highly possible. Thus the expensive and laborious laboratories for the development of microsatellite-flanking PCR primer from genomic libraries of Red Junglefowl are disregarded. Second, there are several technical and analytical advantages of microsatellite marker over other markers for instance; microsatellite alleles are inherited in co-dominant fashion so all alleles can be decriminated. Microsatellite sequences are ubiquity through the genome and selective neutrality. Microsatellite DNA can be amplified by PCR that make it possible to analyze the small amount of DNA. May some microsatellite loci exhibit high polymorphism. Thus this marker are suitable for the determination of various fine parameters in population genetic such as genetic diversity in species that show low level of variation, genetic variations in the intraspecific level, population substructure in recently seperated population.

In this study, six highly polymorphic microsatellite marker that developed within the laboratory of Hans Cheng (Cheng and Critteden, 1994; Cheng et al., 1995), Crooijmans et al. (1997) and Gibbs et al. (1997) were screened. The microsatellite - flanking PCR primers which shown polymorphic amplified products were used to examine microsatellite vriation in genomic DNA extracted from feather or blood stain by using chelex <sup>®</sup>100.

## Objectives

1. To evaluate the possibility of using some chicken microsatellite-flanking PCR primer to investigate the genetic variablility of Red Junglefowl.

2. To use the available domestic chicken microsatellite markers to examine genetic variation within and between the red ear lobe Red Junglefowl inhabits in the northern and southern Thailand.

## Anticipated benefit

To obtain basic knowledge about genetic diversity among the red earlobe Red Junglefowl inhabit in fragmented, geographical different of forests in the northern and southern Thailand. This could be used for interpreting the natural history of *Gallus gallus spediceus* population for providing useful clues about population genetic structure that are valuable in developing an effective management strategies. The wild populations of this species are the important genetic resources in the future for the improvement of native chicken.