Chapter 2

Literature Review

The species was first described by Linnaeus as *Phasianus gallus* in 1766 and changed to *Gallus gallus* in 1758. At least eight other synonyms have been published since then. The subspecies *spadiceus* was described by Bonaterre, possibly in the 1940's ."Junglefowl from Pacific Islands" by Ball (1933) described chicken specimens collected throughout the small islands of the Pacific Ocean. At that period, Um Pang district, Siam now Thailand was listed as one of the places where specimens had been collected. The specimens were also measured for some morphometric characters. A reference cited in this paper (Beebe, 1921) described the estimated distribution of this species, which was said to range from northern India in the southern edge of the Himalayas westward to southern Sikkim, Nepal and Kashmir. Eastward it is commonly found in the hilly portions of Bengal and Assam, throughout Burma including Pegu and Tennessarim, Yunnan, Siam, Cochin China and southward to the Malay Peninsula and Indonesia including Bali, but absent from the Island of Singapore. .

Delacour (1947) noted that the Red Junglefowl is an ancestor of domestic poultry and Beebe (1921) emphasized the constant interchange of blood between wild and domestic birds resulting in variation of habits and coloration unique among pheasants. Smithies (1986) said that the cock in flight is easily recognized by the white patch over the tail. Medway and Wells (1976) noted that the Red junglefowl has been seen in Phuket and Langkawi Islands in Andaman Sea.

Austin Jr. (1963) said that the junglefowl is different from other members of the family Phasianidae in having a comb and wattles about the head, and in having an arched and curved tail.

Ecology and life history

Red Junglefowl are found from sea level to approximately 2000 meters, in tropical and subtropical habitats. The name Junglefowl is a slight misnomer, as they prefer secondary growth to dense primary forest. Forest edge, lightly logged and particularly bamboo forests are all typical habitats in which they are found. (Johnsgard, 1986) Their preferred habitat usually is open forest. There have been no observations of either the bird or its call in deep forest and/ or at high altitude (Collias and Saichuae, 1967). This sexually dimorphic, polygamous species is almost exclusively ground living, flying only to safety, to roost, or when chasing or being chased by another Junglefowl (Sullivan, 1991).

The species feeds on various seeds, fruits, grass, leaves, and insects. Invertebrates from a small but consistent proportion of the diet, particularly caterpillars, termites and dung associated insects (Collias and Saichuae, 1967). Scratching at the ground to find food occupies a large part of the birds' time in the wild.

Red Junglefowl live in flocks with different numbers of cocks and hens. Collias and Saichuae (1967) observed that the sex ratio of flocks ranged from lone males to the groups of 2 males and 6 females.

Brisbin (1969) suggested that there is a behavioral difference of wildness in Red Junglefowls, including wariness, increased in flight distances and a tendency to avoid the presence of man, all of which are greater than in their domestic counterparts. He also suggested that the male red junglefowl always undergoes

eclipse molting after the breeding season. Sexual maturity is reached in the first year, although the males do not develop fully grown spurs and plumage until their second year. The breeding season is from March to September, depending on locality, with 6-12 eggs being laid in nests which are simple scrapes on the ground, hidden in the undergrowth. While incubating her eggs, the hen will occasionally leave the nest very briefly to feed, drink, preen and defecate. After 21 days the eggs hatch and the hen and chicks form a unit independent from her original flock and for the large part away from any males, although males may occasionally consort with these hens. Chick mortality is extremely high since there are many predators including snakes , lizards, birds of prey, and small and large carnivores such as wild cats and civets (Beebe, 1921).

Genetic factors are one of the priorities to be studied in pheasants (Gaston, 1992) since wild Red Junglefowl may be an important source of genetic diversity for future breeding programs especially of domestic *Gallus gallus*. The entire wild gene pool holds enormous potential benefits to the poultry industry.

Mitochondrial DNA of birds

The avian mitocondrial DNA (mtDNA), like that of most eukaryotes, is an extrachromosomal DNA, which is found in the mitochondria, the powerhouse of every cell. It is a single circular molecule, approximately 16000-20000 base pairs long. Unlike nuclear DNA, mtDNA is maternally inherited and undergoes rapid evolutionary change in its nucletide sequence compared to nuclear DNA. The rate of nucleotide substitution on the mitochondrial genome has been estimated to be about 5-10 times more rapid than that of nuclear DNA (Brown, 1979). Mitochondrial DNA is usually monoclonal and does not seem to undergo recombination in vertebrates. Once a variant is established in a female, all

descendants of that individual carry it and, therefore, the inheritance pattern is cloned through the maternal lineage.

Mitochondrial DNA from animals has been well characterized over the past decades. The complete nucleotide sequence of mtDNA of humans (Anderson *et al.*, 1981), cattle (Anderson *et al.*, 1982), mouse (Bibb *et al.*, 1981) and clawed frog (*Xenopus laevis*, Roe *et al.*, 1985) have been reported. These studies reveal that the gene content and genomic organization has remained stable since the divergence of the mammalian and amphibian lineage, approximately 350 million years ago (Brown, 1983). Data from fish mtDNA suggested that a gene content and genomic organization similar to mammals and amphibians exist in fish mtDNA.

The gene order of chicken (*Gallus domesticus*, Desjardin and Morais 1990) in mitochondrial DNA is ND5, cytochrome b, $tRNA^{Thr}$, $tRNA^{Pro}$, ND6, $tRNA^{Glu}$, the control region (D-loop), $tRNA^{Phe}$ and srRNA. This order is identical to that of Snow Goose (Quinn and Wilson, 1993), Japanese quail (*Coturnix japonica*, Desjardin and Morais 1990), and duck mtDNA but differs from that of mammals and frog (*Xenopus*). Within the control region, several short sequences common to mammals are also conserved in birds.

Desjardin and Morais (1990) reported the sequence and gene organization of chicken mitochondrial DNA by cloning and sequencing the whole mitochondrial genome of a domestic chicken, variety white Leghorn. They found that the 16755 base pairs of avian mitochondrial genome encodes the same set of genes (13 protein genes, 2 rRNA genes and 22 tRNA genes) as do other vertebrate mtDNA (Figure 2.1) and is organized in a very similar economical fashion.

Despite these highly conserved features, the chicken mitochondrial genome displays two distinctive characteristics. First, it exhibits a novel gene order, the contiguous $tRNA^{Glu}$ and ND6 genes are located immediately adjacent to the

displacement loop region (D-loop) of the molecule, just ahead of the contiguous $tRNA^{Pro}$, $tRNA^{Thr}$ and cytochrome b genes, which border to the D-loop region in other vertebrate mitochondrial genomes. This unusual order is conserved among all studied galliform birds. Second, a light strand replication origin, equivalent to the conserved sequence found between the $tRNA^{Cys}$ and $tRNA^{Asn}$ genes in all vertebrate mitochondrial genomes thus far, is absent from the chicken genome. These observations indicate that the galliform mitochondrial genome departed from its mammalian and amphibian counterparts during the course of evolution of vertebrate species.

The chicken displacement loop region (D-loop) is delimited on its 3'(prime) end by the gene for $tRNA^{Phe}$ and on its 5' end by the $tRNA^{Glu}$ gene. In the other vertebrate mtDNA sequenced thus far, the 5' end of the D-loop region is bordered by the gene for $tRNA^{Pro}$. This species difference reflects the transposition of the $tRNA^{Glu}$- ND6 mtDNA fragment that has occurred in chicken. (Fig.2.3). The length of the entire control region is the most variable. The D-loop region in chicken mtDNA is slightly larger (1227 bp) than the corresponding sequence found in human (1122 bp), mouse (879 bp), rat (898 bp) or cow (910 bp) mtDNA but is much shorter that of *X. laevis* (2134 bp).

Sequence analysis of vertebrate mtDNA has revealed that the D-loop region is the most rapidly evolving part of the genome. Together with many reasons above, mtDNA is potentially useful for population genetics and molecular systematic studies of animal taxa due to its high mutation rate. Therefore, this study used the mitochondrial D-loop sequence variation as a marker to look at the intraspecific level between two subspecies of Red Junglefowl in Thailand.

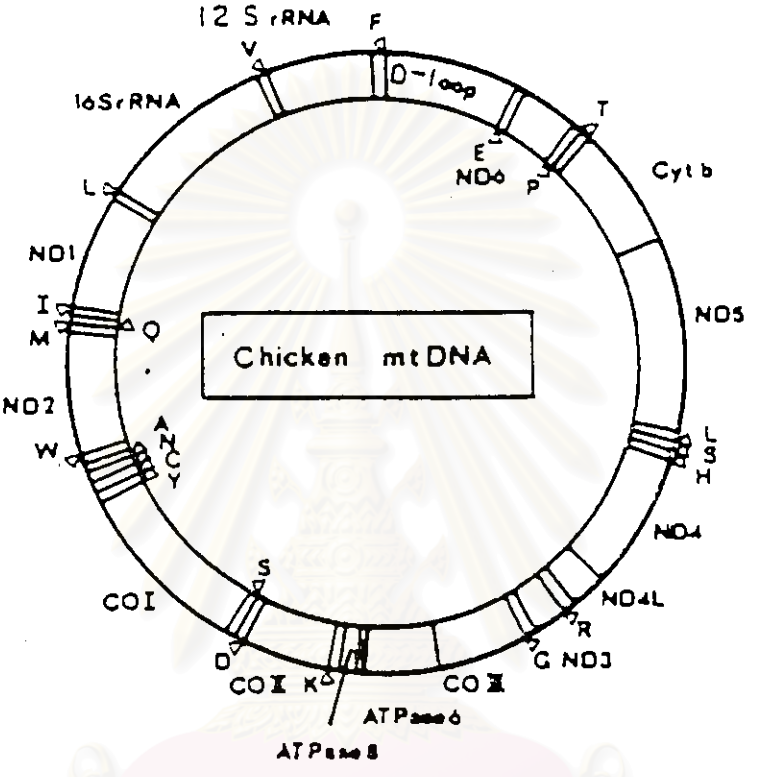

Figure 2.1 Chicken mitochondrial DNA; its genes and gene organization.

(Desjardin and Morais, 1990)

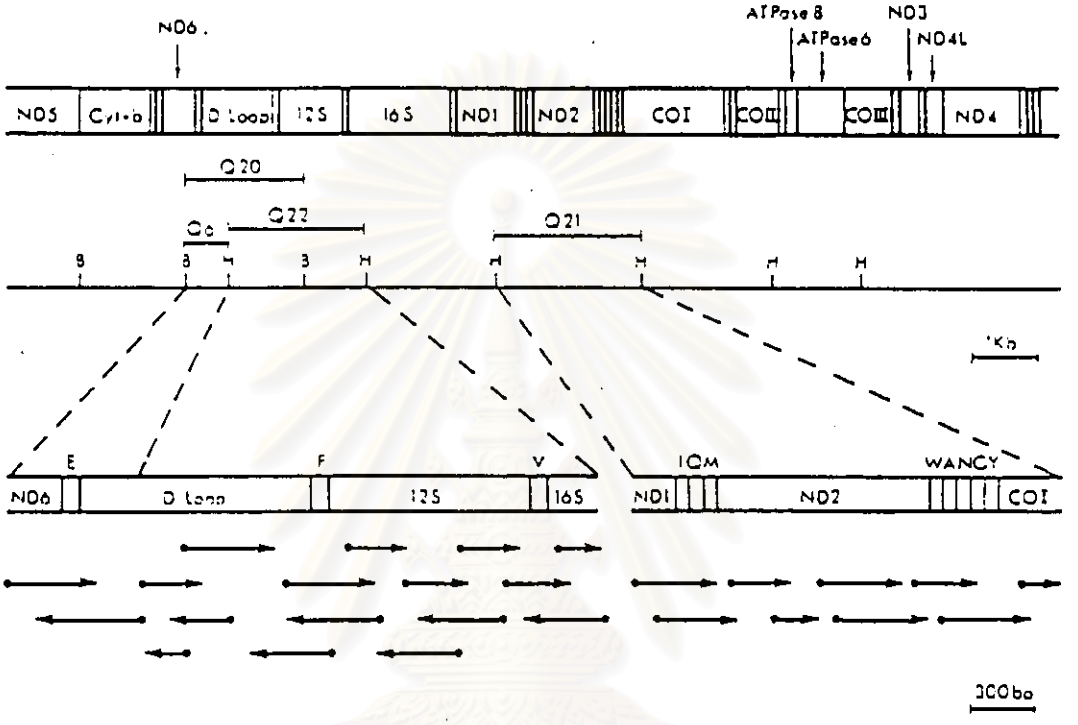

Figure 2.2 A linear presentation of the chicken mitochondrial DNA molecule showing the localization of the genes for cytochrome oxidase subunit I,II and III, (CO I, COII and COIII), ATPase subunit 6 and 8, apocytochrome b (cyt-b), NADH dehydrogenase 1-6 (ND1,2,3,4,4L,5,6), the small (12S) and large (16S) ribosomal DNA subunit, 22 tRNAs (narrow boxes) and the control region (D-loop).

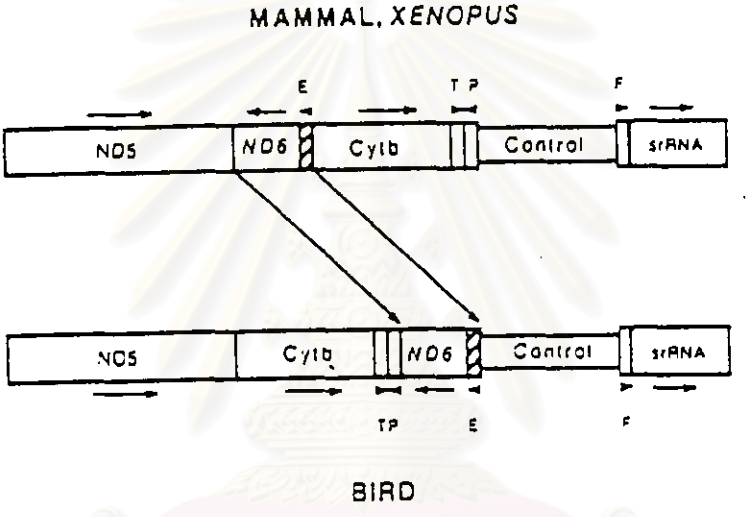

Figure 2.3 Gene order in birds compared to mammals and *Xenopus*. Horizontal arrows indicate the coding strand (to the right, encoded on the heavy strand) Control: control region. (Quinn and Wilson, 1993)