**Doctoral Thesis** 

Studies on Thermoregulation and Growth Performance in Chicks with av-UCP mutation

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### **Summary**

Uncoupling protein (UCP) is a mitochondrial inner membrane protein that is involved with non-shivering heat production. It is protein that produces heat by uncoupling oxidative phosphorylation in mitochondria. In mammals, five UCPs, UCP1, UCP2, UCP3, UCP4, UCP5 have been identified in adipose tissues, muscles, and internal organs. In birds, one UCP (av-UCP) has been found, with a high homology with mammalian UCP3. Av-UCP can influence mitochondrial energy metabolism and heat production. Single nucleotide polymorphism (SNP) of av-UCP have been reported. The SNP locates at exon 3 of av-UCP gene and causes amino acid substitutions. The aim of the present study was to determine the association between the SNP of av-UCP and thermoregulation and growth performance in young chickens.

#### The relationship between thermoregulation behavior and av-UCP gene mutation

Av-UCP is known as a key protein for thermoregulation in poultry and a SNP of the gene has been reported. The purpose of this chapter is to clarify the association between the av-UCP gene mutation and thermoregulation behavior in chickens. Both wild and mutant type chicks for the av-UCP gene SNP (g. 1270; C>T) were exposed to high ambient temperature. We measured rectal temperature, radiation temperature on the body surface and the expression of heat dissipation behavior (wing drooping and panting) during heat exposure. In the measurement of heat dissipation behavior, the latency of intermittent and continuous expression was recorded separately. As a result, changes in radiation temperature on wing during heat exposure in wild type chicks lower than those in mutants. In addition, the latency of continuous wing drooping during heat exposure in wild type chicks was faster than the mutant type chicks. The results from this study suggest that the gene mutation of av-UCP is associated with thermoregulation behavior. However, it is unclear whether the gene mutation affected heat production or heat dissipation.

## *Av-UCP* single nucleotide polymorphism affects heat production during cold exposure in chicks

In previous chapter, we showed the av-UCP mutation associates thermoregulation behavior under high ambient temperature environment. The gene mutation is presumed to affect the heat balance. In this chapter, we exposed the wild and mutant type chicks to a cold environment and clarify the relationship between the av-UCP gene SNP and heat production.

Both the wild and mutant type Rhode Island Red chicks were exposed low temperature (16°C for 15 min) and their physiological responses were compared. The rectal temperature of wild type chicks after cold exposure was higher than those of mutant type. The av-UCP, carnitine palmitoyltransferase-1 expression in pectoral muscle were higher in wild type than mutant type. Hypothalamic gene expression of thyrotropinreleasing hormone and proopiomelanocortin genes were higher in wild chicks than mutant chicks. However, gene expressions of corticotropin-releasing hormone, arginine vasotocin, brain-derived neurotrophic factor and neuropeptide Y did not differ. In plasma metabolites, free fatty acid level in wild type were lower than those of mutant type. Thus, it is suggested that the gene mutation of av-UCP affects non-shivering thermogenesis via the hypothalamo-pituitary-thyroid axis.

# The relationship between mitochondrial, live body oxygen consumption and av-UCP gene mutation

Mitochondria are organelles that synthesize ATP, but they also play an important role in heat production. Heat production in these mitochondria is closely related to respiration, or oxygen consumption. In previous chapter, we showed that the av-UCP gene mutation associates with heat production. Thus, the purpose of this chapter is to clarify the association between the av-UCP gene mutation and mitochondrial oxygen consumption. We isolated mitochondrial from sperm and measured oxygen consumption of the mitochondria. Addition, we measured oxygen consumption of live body. In results, mitochondrial oxygen consumption was higher in wild type than mutant type. In addition to mitochondrial oxygen consumption, we also measured mitochondrial NADH oxidation capacity. The NADH oxidation was higher in wild type than mutant type. In oxygen consumption of live body, that is also higher in wild type than mutant type. These results suggest that the gene polymorphism of av-UCP affects mitochondrial energy metabolism.

### The relationship between growth performance and av-UCP gene mutation

In chickens, a single nucleotide polymorphism (SNP) of the av-UCP (avian UCP) gene has been reported to be associated with body weight gain and increased abdominal fat. As previous chapters, the gene mutation of av-UCP affect heat production in chicken. Heat production is an indicator that is deeply linked to chicken productivity or growth. Therefore, UCP exists in the inner mitochondrial membrane and affects the gradient of the electron transport chain and proton pump, so it is expected to affect body composition. The objective of this study was to evaluate the relationship between body composition

and av-UCP gene mutation. In the results of free amino acids concentrations in pectoral muscle, the effect of sex was observed at Threonine, Glutamine, Lysine and Arginine. However, the effect of av-UCP mutation and the interaction between sex and the mutation were not observed in any results. As a result of chemical analysis of the whole carcass, no effect of gene polymorphism was observed on the contents of moisture, crude protein, crude fat and crude ash. In body weight at 8 weeks old, there was a tendency for the effects of gene polymorphisms to be observed, and the wild type was larger than the mutant type. It has been suggested that UCP affects the gradient of the mitochondrial electron transport chain, the proton pump. Therefore, it is inferred that UCP affects the acid-base balance in cells. Given these possible effects of UCP, we hypothesized that gene mutation on the UCP might alter the body composition of chicken. In addition to these, it has been reported that UCP gene mutations affect body shape in small mammals such as mice and rats, and human. And more, it has been reported that the gene mutations targeted in this study also affect chicken body weight gain and intra-abdominal fat. However, in this study, gene polymorphisms did not affect body composition. It has been reported that the effect of this gene mutation on performance varies depending on the chicken lines. Therefore, it is necessary to investigate chickens of other strains as well.

### Conclusion

In conclusion, av-UCP gene mutation affect the mechanism of heat production in mitochondria. Also, since this mutation does not affect muscle amino acid composition, it does not affect productivity. However, although it has been reported that this gene mutation affects body gain performance, it has been suggested that the effect differs depending on the chicken breed. Therefore, it is effective to use this gene mutation for breeding to acquire heat tolerance, but it is necessary to consider it depending on the breed.