

Single Gene Consistently Associated with Heat Stress Response in Three Distinct Chicken Lines

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Summary and Implications

Identification of specific gene or quantitative trait locus that responds to specific stimulus is an important goal for genetic improvement programs. In a heat stress experiment with 3 distinct chicken lines, we found Angiopoietin-like 4 (ANGPTL4) to be differentially expressed in all 3 lines during acute and chronic heat stress. This outcome suggests the need to further explore ANGPTL4 as a potential candidate gene for selection to improve chicken response to heat stress.

Introduction

Heat stress has a significant negative impact on the poultry industry through economic losses of mortality and decreased performance. In this experiment, 3 distinct genetic lines maintained at Iowa State University were utilized: a heat-susceptible broiler line, a heat-resistant inbred Fayoumi line, and the F₁₉ generation of a highly advanced intercross line (AIL) between the broiler and Fayoumi. Comparing the RNA expression from liver samples procured during a heat challenge experiment, we aimed to identify differences in gene expression as measured by RNA sequencing.

Materials and Methods

An equal number (n=16) of 3-week old male chicks from the 3 genetic lines were used (n=48). The chicks were divided equally into 2 groups: 1) the heat stress group (n=24) that was subjected to a cyclic heat stress of elevated temperature (35°C) for 7 hr and normal temperature (25°C) for 17 hr, and 2) the contemporary control group (n=24) that was kept at the normal temperature (25°C). Birds in the acute heat treatment (n=24, 12 per group) were euthanized

on day 1 after 3h of heat exposure in the heat stress group. Chronic heat treatment accounts for the other half of the birds (n=24, 12 per group) that were euthanized on day 7 of heat exposure in the heat stress group. Liver samples were processed for RNA sequencing on the Illumina HiSeq 2500. The resultant sequencing reads were mapped to the chicken reference genome (*Gallus gallus* 4.0), and edgeR identified differentially expressed genes (DEGs) by comparing the gene expression differences between the heat stress group and the control group for both treatments.

Results and Discussion

The acute heat treatment resulted in 627 DEGs in broilers, 78 DEGs in Fayoumi, and 128 DEGs in AIL with 9 DEGs shared between the 3 lines (Figure 1A). The chronic heat treatment resulted in 129 DEGs in broilers, 163 DEGs in Fayoumi, and 86 DEGs in AIL with 5 DEGs shared between the 3 lines (Figure 1B). The only gene shared between all 6 sets of DEGs was ANGPTL4, a key protein involved with energy homeostasis and induces nitric oxide production during wound healing. However, the role of ANGPTL4 has not been well characterized in chickens.

Our experiment shows that there were differences in expression pattern for ANGPTL4 among the 3 lines (Table 1). Broilers had reduced expression of ANGPTL4 for both treatments; in contrast, Fayoumi had increased expression of ANGPTL4. The AIL showed a mixed response with an expression pattern similar to broilers during acute heat treatment but an expression pattern similar to Fayoumi during chronic heat treatment. As a combination of both broiler and Fayoumi lines, it is logical that the AIL response resembles aspects of both its parent lines. Given the differences in expression patterns for ANGPTL4 among the lines of chickens that have differences in susceptibility to heat stress, ANGPTL4 may be a gene of interest to be further explored for genetic improvement of resilience to heat stress in chickens.

Acknowledgments

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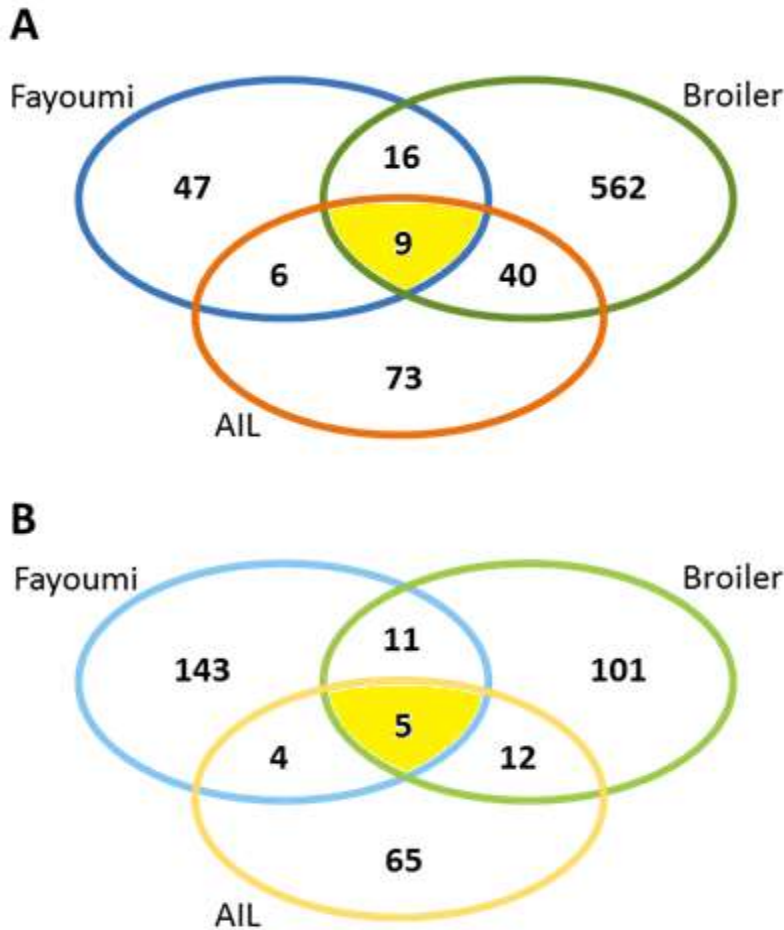


Figure 1. Differentially expressed genes (DEGs) results. (A) Venn diagram of the acute heat treatment vs. control DEGs for all 3 chicken lines. There are a total of 9 genes found in all 3 contrasts. (B) Venn diagram of the chronic heat treatment vs. control DEGs for all 3 chicken lines. There are a total of 5 genes found in all 3 contrasts. The common intersection among the three lines in both Venn diagrams contains ANGPTL4 and is highlighted in yellow.

Table 1. Expression pattern of ANGPTL4.

Genetic lines	Acute Heat Stress	Chronic Heat Stress
Broiler	↓	↓
Fayoumi	↑	↑
AIL	↓	↑