Heat Stress Alters Immune Pathways in Liver of Divergent Chicken Lines

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Summary and Implications

The liver plays a variety of roles in energy metabolism, digestion, and immune response. During the challenge by an environmental stressor, such as heat, the liver is one of the key organs that determines the chicken's ability to cope with the hostile environment. Sequencing RNA extracted from liver samples from a heat-susceptible broiler line and a heat-resistant Fayoumi line, we have identified a set of genes that were differentially expressed due to an acute heat stress challenge. Analysis of these genes in context of the biological pathways showed opposite responses by the broiler and Fayoumi chickens, but there was activation of immune signaling pathways for both lines. This result suggests that despite mechanistic differences in heat stress response, selection for improvement in heat tolerance may also cause alteration to immune response.

Introduction

As we continue to deal with the impact from climate change, the increased frequency of heat waves poses a challenge for the poultry industry worldwide. Past selection efforts for improved meat production in broilers also resulted in increased susceptibility to stressful environments, such as heat stress. One of the key organs in the chicken's response to heat stress is the liver. Comparing liver samples from a broiler line to liver samples from an inbred more heat-tolerant Fayoumi line that originated from Egypt, we hypothesize differences in biological pathways as measured by differential gene expression based on RNA sequencing. Improved understanding of the mechanistic differences in heat stress response in these two distinct lines of chickens will assist in the future of breeding more heat-resistant chickens.

Materials and Methods

A total of 16, 3-week old male chicks – 8 broilers and 8 Fayoumis were used in this study. The chicks were separated into the heat stress group (n=8, 4 per line) that was exposed to high ambient heat of 35°C and the control group (n=8, 4 per line) that was maintained at 25°C. Liver samples from all 16 birds were collected at 3h into the thermal treatments and 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 differentially expressed genes were identified with edgeR software. These genes were loaded into Ingenuity Pathway Analysis (IPA) software for biological pathway analysis.

Results and Discussion

Analysis in IPA for broilers found 13 biological pathways that were enriched in the heat-stressed birds, and all 13 pathways were activated with the exception of PTEN signaling (Figure 1A). These activated pathways are predicted to impact several essential biological systems: cardiovascular, nervous system, immune system, hepatic system, and circulatory system. The analysis for the heat-stressed Fayoumi found 10 enriched pathways, but 7 of 10 enriched pathways were down-regulated, suggesting a very different response to heat stress by the Fayoumis compared to the broilers (Figure 1B). Of the 3 activated enriched pathways in Fayoumis, all were associated with the immune system. In response to the acute heat stress, the two chicken lines had opposite responses in biological pathways suggesting a potential explanation for the difference in susceptibility to heat stress. However, both chicken lines had activation of immune signaling pathways, suggesting that selection for improvement in heat stress response may have the added effect of increasing immune response.

Acknowledgments

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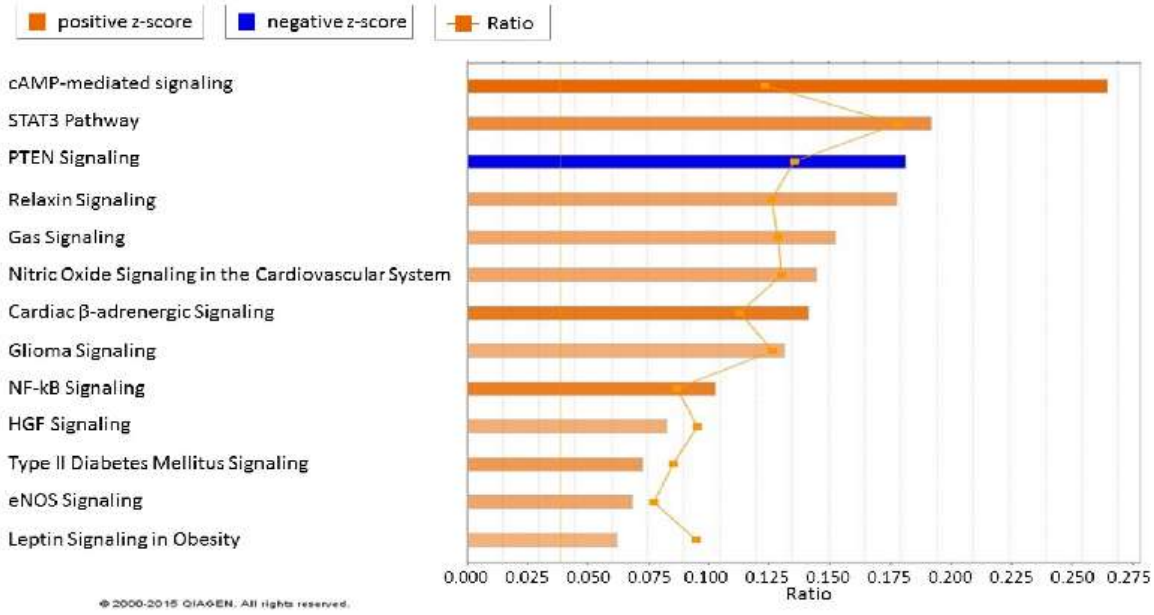
A



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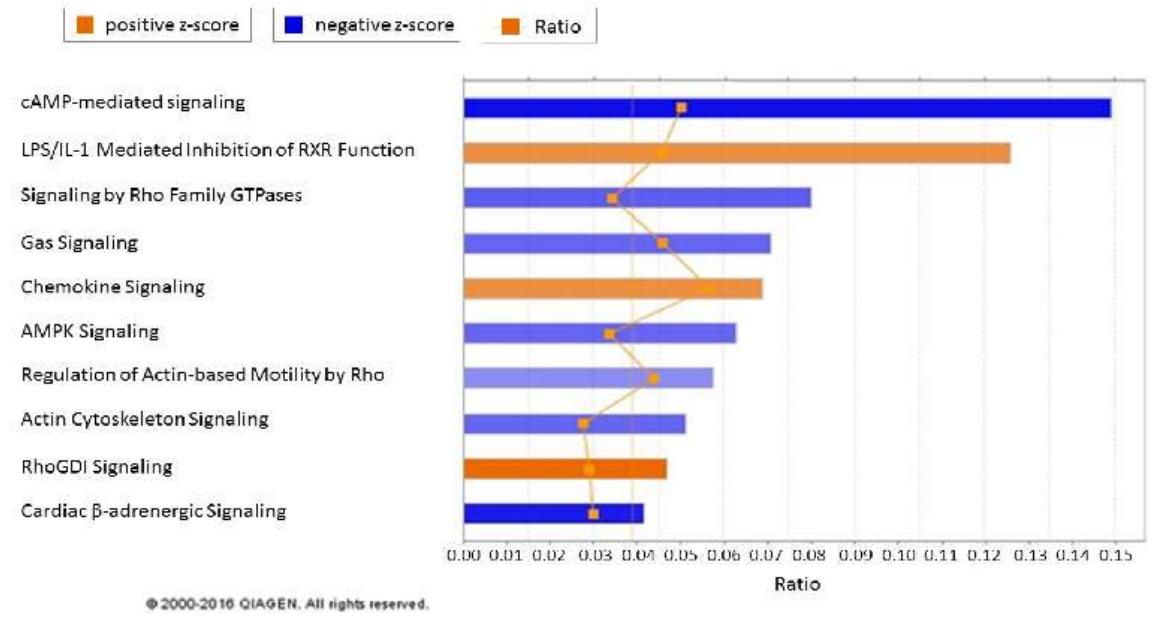

Figure 1. IPA functional analysis of biological pathways. The biological pathways with the most significant change in acute heat stress are shown for: (A) broiler acute heat stress group compared to control group showed activation of several biological systems: cardiovascular system (Nitric Oxide Signaling in the Cardiovascular System and Cardiac β-adrenergic Signaling), nervous system (Glioma Signaling), immune system (NF-kB Signaling), hepatic system (HGF Signaling and Leptin Signaling in Obesity), and circulatory system (eNOS Signaling). (B) Fayoumi acute heat stress group compared to broiler acute heat stress group showed down-regulation of most pathways except for the activation of immune system (LPS/IL-1 Mediated Inhibition of RXR Function, Chemokine Signaling, and RhoGDI Signaling). The more orange the bar in the chart, the greater the activity. In contrast, the more blue the bar in the chart, the greater the inhibition.