Cecal Microbiome Characterization for Layers under Heat Stress

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Summary and Implications

Animal microbiomes have gained attention in recent years because of their roles in a variety of physiological responses to disease and environment challenges. In this study, we subjected production white egg layers to a 4-week heat stress challenge to measure changes in the cecal microbiome. We found that heat stress alters the cecal microbial composition after 2 weeks of cyclic heat exposure. To our knowledge, this is the first study to demonstrate changes in microbiome in heat-stressed layers, and provides an approximate time course for the microbiome alterations.

Introduction

One response from chickens under heat stress is decreased feed intake that negatively impacts performance traits. Although prior studies have shown changes in cecal bacterial composition in broilers during heat stress, the impact of heat stress on the layer microbiome has yet to be demonstrated. By treating a group of laying hens to cyclic ambient heat stress, we emulate a heat wave that is typical for U.S. layer production. Collecting and analyzing the cecal microbiome, we hope to elucidate the changes that occurs for the microbiome during heat stress.

Materials and Methods

White-egg layers (n=80) were randomly assigned to heated or control groups. The heated group was exposed to a daily cyclic heat stress of elevated temperature (35°C) for 7 hr and normal temperature (28°C) for 17 hr. The control group was held at a constant temperature (21°C). Cecal content was collected from 8 layers from each of the 2 treatment groups at 5 time points: 3h, 1wk, 2wk, 3wk, and 4wk. The samples were flash frozen, transferred to the lab, and extracted DNA was processed for 16S rRNA amplification and sequencing by Illumina MiSeq. The sequences were uploaded to MG-RAST for analysis, where relative abundance profile and alpha-diversity were calculated for each sample.

Results and Discussion

The relative bacterial abundance profiles showed that the heat stress and control groups had very similar microbial compositions at the start of the experiment (3h) and *Firmicutes* was the most abundant phylum followed by *Bacteroidetes* (Figure 1). However, the two groups began to exhibit differences in these two phyla as the experiment continued, with the biggest difference in the bacterial composition after 4 weeks. The alpha-diversity (an estimate of bacterial species presence) shows a clearer picture of the differences in microbiome between the two groups (Figure 2). The heat group had an increase in bacterial species present starting at 2 weeks and continued to have greater diversity compared to the control group until 4 weeks. The alpha-diversity suggests that the heat group underwent a disturbance of the microbial community after 2 weeks of heat stress and returned to the baseline level of bacterial species richness after the restructuring at 4 weeks. From this characterization of the microbiome, we observed that it takes at least 2 weeks before there was a measurable change in the cecal microbiome. The data also suggest that heat stress leads to a restructuring of the cecal microbial population that is suited for the hot environment by 4 weeks.

Acknowledgments
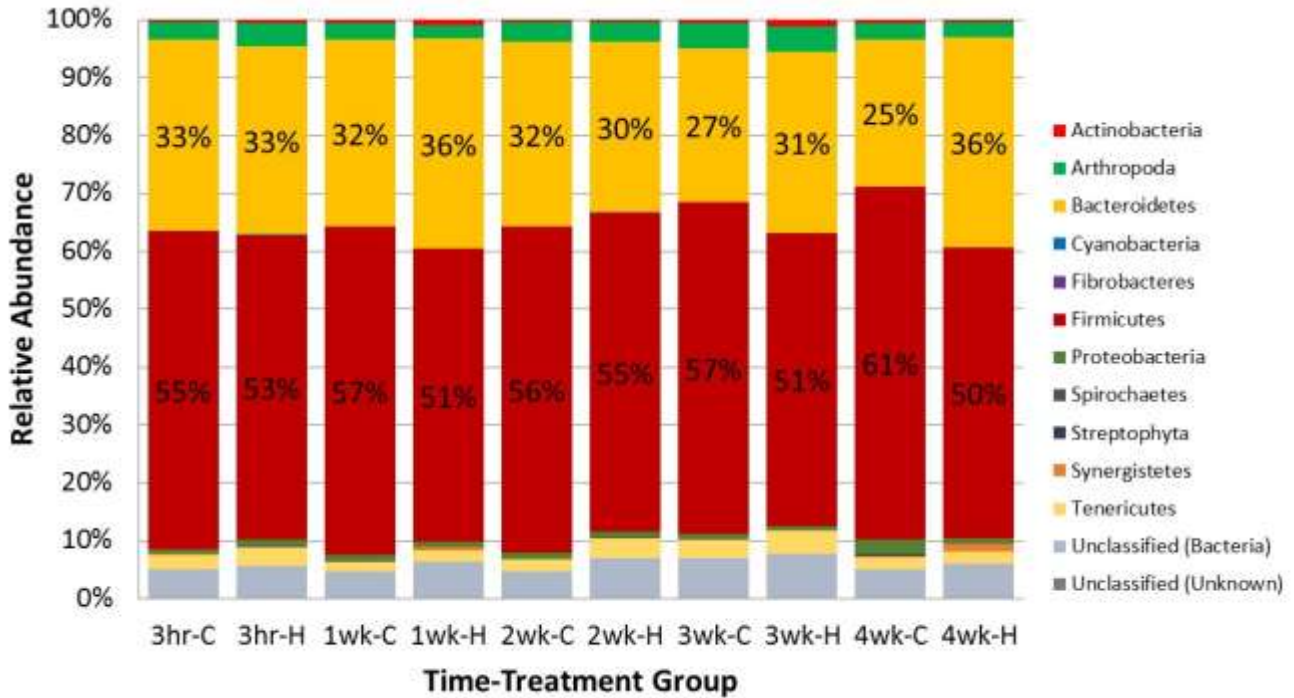
Figure 1. Relative phyla abundance of layer cecal microbiome of heat stress and control groups. Phyla are colored according to the key on the right, of note, red = *Firmicutes* and yellow = *Bacteroidetes*, which are the two most abundant phyla. C = control group and H = heat stress group
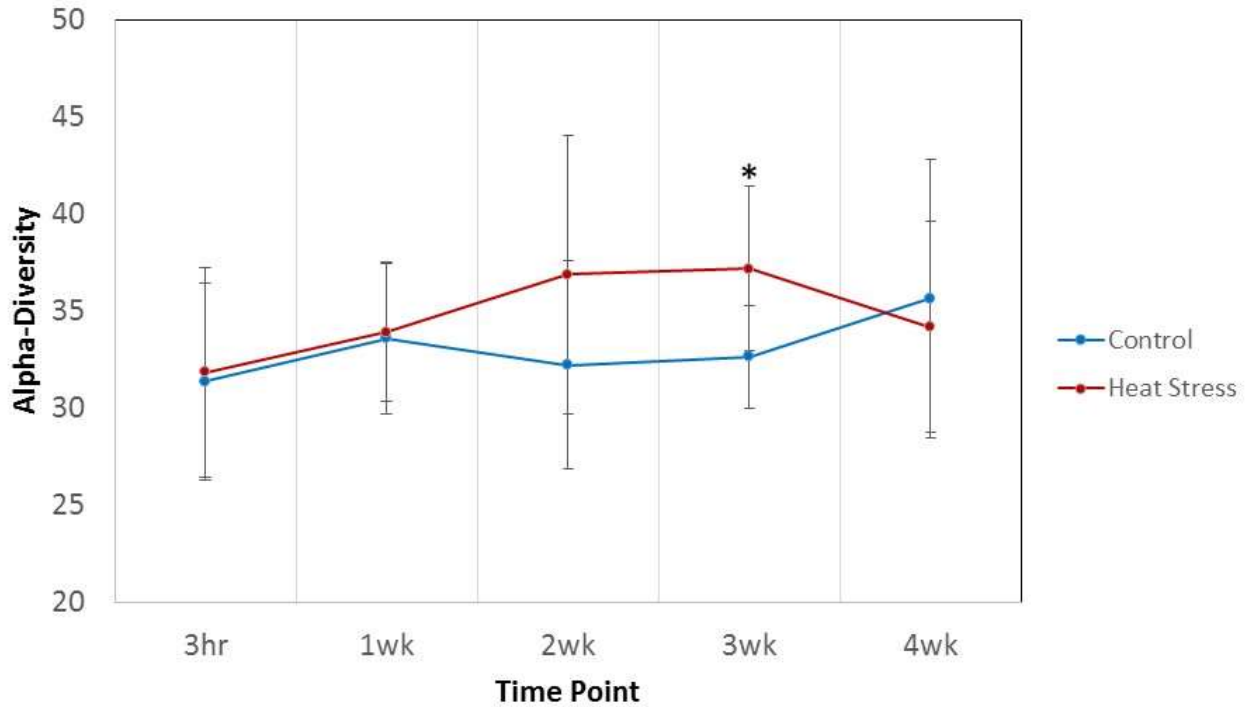
Figure 2. Alpha-diversity of heat stress and control groups. Alpha-diversity is an estimate of bacterial species present. The blue line represents the heat stress group and the red line represents the control group. *There was a significant difference ($P<0.05$) in the alpha-diversity at 3 weeks.