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Laying traits and underlying transcripts, expressed in the hypothalamus and pituitary gland, that were associated with egg production variability in chickens

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Abstract

The objective was to characterize the potential laying traits and underlying transcripts expressed in the hypothalamus and pituitary gland that were associated with egg production variability in five genetic stocks of chickens: two commercial lines, Red- ($n = 12$) and Black-feather ($n = 14$) Taiwan country chickens (TCCs); two selected lines of TCCs, B (high body weight/comb size; $n = 17$) and L2 (high-egg production; $n = 14$); and a commercial single comb White Leghorn (WL; $n = 17$). Six laying traits, age at first egg, clutch length, pause length, oviposition lag within clutch, follicle rapid growth period, and rate of yolk accumulation were measured. The significance of differential values among five chicken stocks and correlation coefficients between laying traits and number of eggs to 50 weeks of age or laying rate after first egg, and the expression level of 33 transcripts were determined. Longer clutch length and shorter oviposition lag within clutch contributed to a higher number of eggs to 50 weeks of age or laying rate after first egg in L2 ($P < 0.05$) and WL strains ($P < 0.05$). However, their rate of yolk accumulation ($P < 0.05$) and follicle rapid growth period ($P < 0.05$) were different, indicating the accumulation of different alleles after long-term, independent selection. Across all five strains, numbers of eggs to 50 weeks of age were positive correlated with average clutch length ($P < 0.05$) as well as the rate of yolk accumulation ($P < 0.05$). Expressions of *PLAG1*, *STMN2*, *PGDS*, *PARK7*, *ANP32A*, *PCDHA@*, *SCG2*, *BDH* and *SAR1A* transcripts contributed to number of eggs to 50 weeks of age ($P < 0.05$) or laying rate after first egg ($P < 0.05$). Analysis of correlation coefficients indicated that *PLAG1* additionally played roles in decreasing average pause length. Two transcripts, *PRL* and *GARNL1*, specifically contributed to number of eggs to 50 weeks of age or laying rate after first egg by reducing oviposition lag within clutch ($P < 0.05$) and/or increasing average clutch length ($P < 0.05$), respectively. Expression level of *NCAM1*, contributed to laying rate after first egg by association with a shorter oviposition lag within clutch ($P < 0.05$). The current study attributed egg production phenotype in five strains into several laying traits; correlations between these traits and expression levels of underlying transcripts expressed in the hypothalamus and pituitary gland were also established.

Keywords: Chickens; Clutch length; Egg production; Laying traits; Oviposition lag within clutch; Transcripts

1. Introduction

The Taiwan country chicken (TCC) is a slow growing breed [1], but an important source of both

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meat and eggs [2]. However, the lack of ideal mating systems in the production of most commercial TCC strains has resulted in the inefficient application of heterosis. Selection for part- or whole-year number of eggs or for laying rate is a general approach to improve egg production in hens that yielded some positive genetic progress [3]. To identify factors associated with the variability of egg production, genetic studies have focused on the time interval between consecutive ovipositions [4,5], the mean time of oviposition [6,7], and the intra-clutch mean lag of oviposition time [8], in which the increase in egg production was attributed to an increase in the rate of passage of eggs through the oviduct. Zakaria et al. [9] demonstrated that the number of growing follicles and the total amount of daily yolk deposition in layers with short clutches were less than those exhibiting long clutches. Increased clutch size decreased the average follicle growth period, and increased follicle growth rate. To dissect egg production (a complex phenotype) into laying traits should help to elucidate specific factors affecting egg production variability.

In poultry, similar to other vertebrates (including nondomestic birds), reproductive activities are strictly controlled by the hypothalamic–pituitary–gonadal axis [10]. Minute differences in hypothalamic or pituitary functions, or both, might affect reproductive activities, including folliculogenesis, ovulation, oviposition, and incubation behavior [11]. Consequently, the hypothalamus and pituitary gland are ideal tissues to search for molecular markers associated with egg production, since genes may be transcribed differently under different physiological conditions, i.e., remote or immediate preparations, or both, for egg laying. In previous transcriptomic analysis of hypothalamic and pituitary proteins in TCC strains, B and L2, selected for body weight/comb size and high-egg production for 20 generations, respectively, 53 known and 65 unknown transcripts were identified as candidate transcripts related to high-egg production [12]. Expression levels of nine and 25 in 33 randomly selected transcripts, have been further confirmed to be significantly increased in hens with higher egg production [12,13]. Namely, in seven chicken populations across five genetic stocks, the mRNA expression levels of *BDH*, *PCDHA@*, *PGDS*, *PLAG1*, *PRL*, *SAR1A* and *SCG2* were significantly correlated with number of eggs to 40 weeks of age.

Egg production is a composite phenotype which might be influenced by several laying traits. Therefore, the objective of this study was to identify potential factors and underlying transcripts expressed in the hypothalamus and pituitary gland that affected egg production variability across five different chicken strains. The significance of the differential trait values and the correlation between specific laying trait and the expression level of each candidate transcript were measured.

2. Materials and methods

2.1. Birds

Animal care and usage were reviewed and approved by the Ethics Committee of Laboratory Animals at the National Chung-Hsing University (NCHU), Taiwan. A total of 78 laying hens from five genetic stocks were used in this study. These included two (Black- and Red-feather) commercial TCC strains, two (B and L2) selected TCC strains that were established from a common base population in NCHU since 1982, as well as a commercial stock of single comb White Leghorn (WL). The Black- and Red-feather TCCs are, respectively, the most popular medium and large commercial birds in Taiwan, whereas the B and L2 strains were selected for body weight/comb size and egg production for 20 generations [14]. In order to control the environment as fixed effect, at 14 weeks of age, 18 pullets from each genetic stock were randomly assigned to individual laying cages in a windowless environmental-control facility. A breeder diet in pellet form (100 g/bird, CP 15.0%, ME 2930 kcal/kg, Ca 3.24%) was given (08:00–09:00) to each pullet to the age at first egg; thereafter, the layer diet (110 g/bird, CP 16.9%, ME 2930 kcal/kg, Ca 3.24%) was given to the end of the experiment. Artificial light was provided for 10 h (10L:14D) until the 5% hen-housed egg production rate was reached. Thereafter, the daily photoperiod was extended to 14 h (14L:10D), and then was increased by 1 h/weeks to 16 h (16L:8D) to stimulate ovulation [15] until the end of experiment. Egg production related traits including age at first egg, laying rate after first egg [%; number of eggs to 50 weeks (350 days) of age/(350 − age of the first egg) × 100%], number of eggs to 50 weeks of age, clutch length, and pause length were recorded for each bird from the day of first egg to 50 weeks of age. Deformities, cracks and double-yolk were included in the number of eggs. The clutch length was the number of eggs laid on consecutive days, an important component of the total number of eggs laid during a production cycle. Conversely, the pause length was the interval between two clutches. The clutch and pause length of each population were calculated as the arithmetic mean of all clutches recorded. A few hens in