Genome-Wide Association Analysis Identifies Loci That Influence Ascites In broilers

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Introduction

Ascites (Pulmonary Hypertension Syndrome) is a metabolic disease in broilers and causes mortality of up to 8% in commercial broilers flocks (Maxwell and Robertson, 1998). Ascites is believed to be caused by an imbalance between oxygen requirement and the cardiovascular ability to supply oxygen (Julian *et al.*, 1987). It has been suggested that the incidence of ascites has increased through the years as a consequence of selection for higher meat yield, increased growth rate and lower feed conversion ratio (Balog *et al.*, 2003, Havenstein *et al.*, 2003). Although extensive research has been performed, the cause of ascites and the association to body weight remains unclear (Decuypere *et al.*, 2000).

Identification of quantitative trait loci (QTL) in broilers has been given much attention in recent years. The use of QTL information of ascites susceptibility has been suggested to effectively in controlling ascites susceptibility (Pakdel *et al.*, 2005a). Rabie et al. (2005) performed a linked analysis to find QTL using microsatellites involved in ascites, and found statistical evidence for QTL on several different chromosomes. The aim of this present study was to perform a whole-genome scan with single-nucleotide polymorphism (SNP) used to detect QTLs controlling ascites-related trait, and find possible association between ascites related trait and body weight at two weeks.

Material and methods

Animals. The chickens used in the present study were from a dam line originating from the White Plymouth Rock breed. The experimental population used to detect ascites QTLs was based on three generations (G1, G2 and G3), genotypic information were from G2 and phenotypic information were from G3 were used in the present study. The G2 generation consisted of 891 broilers and G3 generation consisted of 8,158 broilers.

Genotype: In G2 broilers were genotyped for 19,314 SNPs covering the whole genome. Genotyping of SNPs was carried out using Illumina Infinium iSelect Beadchip. Markers on the SNP chip were evenly distributed across the broiler genome, with a marker density of approximately six markers per centimorgan (cM).

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Phenotype. G3 were weighed at two weeks (BW_2). The ratio of the weight of the right ventricle as a percentage of the total ventricle weight (RATIO) was determined for each individual.

Statistical analyses. No phenotypic observations were recorded on G2 broilers, so the phenotypic data of their offspring G3 broilers were used to calculate average adjusted found progeny means for G2 parents as described by van Kaam et al. (1998). The model used to analyze the data was:

$$y_{ijk} = SNP_i + HS - Family_j + e_{ijk}$$

Where y_{ijk} represent the average adjusted trait value of individual ijk, with SNP genotype i, from paternal half sib family j. SNP_i is the fixed effect of the SNP genotype, either AA, AB or BB; HS-$family_j$ is fixed effect of paternal half sib family (j = 1,2,...,69) and e_{ijk} is random residual effect with $e \sim N(0, I_e^2)$.

Results and discussion

Ascites in broilers is a complex disorder. Several traits have been found as indicator traits for the disorder, such as an enlargement of the right ventricle (RATIO), fluid accumulation in the abdomen, weakness of the internal organs, lower BW and eventually the death of the sick broiler (Rabie et al., 2005). In the present study was a whole-genome scan with SNP performed to detect QTLs RATIO, and find possible association between RATIO and BW_2.

RATIO:
The SNPs used in this study were located on the chromosomes 1 to 28 of the chicken genome. Significant test results were found in analysis of the trait RATIO (figure 1). The whole genome-wide association study for ascites in broilers identified 67 significant QTL affecting RATIO located on across the genome. The chicken genome consists of 39 pairs of chromosomes, which means that in the present study 11 chromosomes have not been covered. These 11 chromosomes constitute the smallest of the micro chromosomes and the sex chromosomes in chicken and probably account for <10% of the chicken genome (Rabie et al., 2005).

There was statistical evidence for QTLs for RATIO as an indicator trait for ascites on chromosomes 1, 2, 3, 4, 5, 6, 7, 8, 10, 12, 17, 18, 19, 20, 21, 22 and 28. The most significant SNPs were located on chromosome 12, 18 and 22. Out of the 67 significant SNPs, 59 SNPs had minor allele frequency above 5% and taking multiple testing into account by applying a False Discovery Rate (FDR) had a value above 0.01.

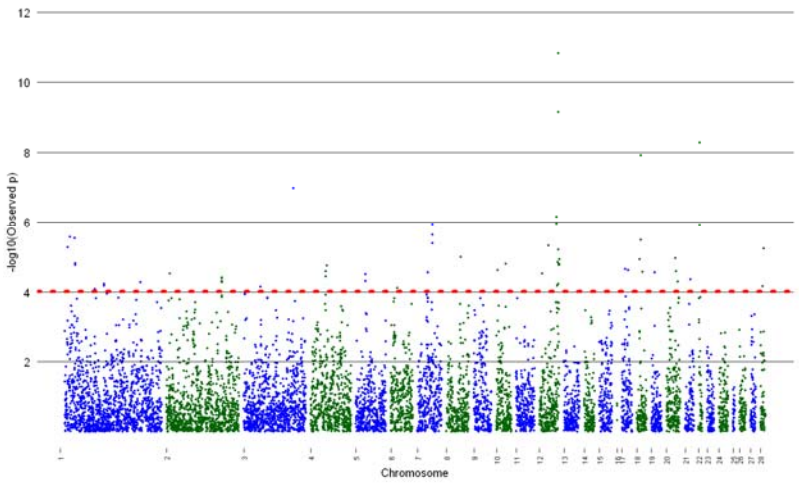

Figure 1 Manhattan plot for the SNPs from the genome wide association. The dotted line represent the FDR on 0.01 and $-\log_{10}(p)$ above 4.0.

RATIO and BW_2: The SNPs with significant and suggestive association for both RATIO and BW_2 are summarized in Table 1. Significant SNPs were located on chromosomes 1, 2, 7, 12, 17, 22 and 28. These results of significant SNPs found for both RATIO and BW_2, indicate that QTLs are located on same regions for both RATIO and BW_2.

Table 1: Significant SNPs for both RATIO and BW ($-\log_{10}$(p value)>4)

SNP	Chromosome	RATIO	BW2
Gga_rs15318316	1	4.18	4.96
Gga_rs14846971	1	4.23	4.97
Gga_rs15876453	2	4.53	4.17
Gga_rs13738250	7	4.56	4.54
Gga_rs14049226	12	10.83	5.37
Gga_rs14986485	12	9.14	5.35
Gga_rs14985701	12	4.17	5.19
Gga_rs14100447	17	4.63	5.34
Gga_rs16183608	22	8.28	4.13
Gga_rs14307070	28	5.25	4.80

The significant SNPs found for both RATIO and BW_2 is supported by the genetic correlations found other studies where genetic parameters for RATIO and BW have been analyzed. Pakdel et al. (2005b) and Closter et al. (2009) found a negative genetic correlation

between BW_5 and RATIO, where Closter et al. (2009) found ac positive genetic correlation between RATIO and BW_2 and negative genetic correlation between RATIO and BW_5.

Conclusion

In this present study we have located significant QTL for the ascites indicator trait, RATIO across the whole genome. There were also found QTLs that were significant for both RATIO and BW_2, and suggest that there is an association between the development of ascites and body weight.

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