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Identification of mutations in a QTL region on chromosome 3 associated with fatness in chickens

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Abstract:

The Brazilian chicken meat production has a great importance in worldwide economy, which is a result of constant improvement of performance traits, mainly due to genetic improvement. Next-generation sequencing (NGS) enables the generation of a larger number of data and the identification of many mutations such as SNPs and insertions/deletions (INDELs). The objective of this st udy was to sequence by NGS with high sequence coverage six individuals from two lines of the EMBRAPA F2 Chicken Resource Population to identify SNPs and INDELs in a QTL region of the chicken chromosome 3: 33,595,706-42,632,651 bp (between LEI0161-ADL037) associated previously with fat deposition (Campos et al., 2009). Three animals of the broiler line TT and three of the layer line CC were sequenced by Illumina sequencer HiSeq 1000 with the initial coverage of 18X/chicken. The library sequencing was prepared with Nextera DNA Sample Preparation kit and the cluster generation with Cbot (Illumina) using TruSeq PE cluster kit V3 (Illumina). The quality trimming was performed to remove reads with low quality (quality >24 and fragment size > 65 bp) by Segyclean (v.1.3.12). Reads were aligned against the reference genome (Gallus gallus-4.0) with Bowtie 2 (v.2.1.0). From the alignment file SNPs/INDELs were identified by SAMtools software (v.0.1.19) with mapping and base qualities. After the SNPs/INDELs discovery, the filtration based on quality parameters (q≥30) and total coverage (DP4≥5) was performed. After the quality trimming, approximately 77.9% of the reads were kept from TT chickens (≅ 170 million reads/chicken). For CC chickens were kept approximately 77.4% of the reads (≅ 140 million reads/chicken). In the target region of GGA3 were initially identified 60,112 SNPs and 5,874 INDELs in the two lines. After filtration were retained 55,778 SNPs and 4,371 INDELs. We removed 7.3% of SNPs initially identified and 25.9% of INDELs. After the filtration, broilers had a larger number of SNPs/INDELs in comparison to the layer line for the target region. The three broilers presented 9.6% more SNPs and 13.3% INDELs than the layers. The largest number of mutations identified in TT line in the QTL region, previously associated with fatness, highlights the importance of studying these variants considering the heritability and variability between the two lines for fat deposition. The MAF was calculated and we identified 28,450 SNPs and 2,731 INDELs fixed and 24,528 SNPs and 1,330 INDELs segregating in the layer line, and 32,279 SNPs and 3,191 INDELs fixed and 26,300 SNPs and 1,491 INDELs segregating in the broiler line. The largest number of mutations segregating in broiler line TT indicates greater variability and consequent variation in fat deposition in the carcass. The SNPs/INDELs found in this study can be used to narrow down the QTL region in the search for the causal mutation.