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## Detection of QTL Influencing Egg Quality Traits in Layers Receiving Various Diets

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**ABSTRACT:** Taking advantage of the 600K Affymetrix® Axiom® HD genotyping array, the aim of this study is to detect QTL influencing egg quality traits in layers. A population of 438 sires from a pure line was genotyped. Their 31,381 F1 crossbred daughters were phenotyped for egg quality traits at 50 and 70 weeks of age. Moreover, to observe the putative existence of QTL by environment interactions, these layers were divided into 2 groups, fed with two different diets. The egg shell color and its components (redness, yellowness, lightness), the egg shell strength, stiffness and shape, the short length of egg and the egg weight were recorded. GWAS analyses have been showed 13,547 SNP with a significant (1% chromosome wide level) association with at least one egg quality trait. Thus, QTL have been detected and genotype by environment interactions have been observed.

**Keywords:** laying hens; GWAS; QTL

### Introduction

Since decades, traits selected in layers concern egg number and egg quality performances. Even if these traits were improved by selection, their genetic architecture, i.e. the quantitative trait loci (QTL) influencing traits, is still poorly known. As of today, 33 studies focused on QTL detection for egg production in chicken (Chicken QTLdb) and only few genes were identified as involved in egg number or egg quality determinism.

Recently, the development of a high density chip for chicken by Affymetrix, i.e. the 600K Affymetrix® Axiom® HD genotyping array (Kranis et al. (2013)), allows envisaging the use of high density genotype data for genomic selection in laying hens. This chip could also allow narrowing the localization of QTL previously detected and to detect new QTL involved in egg production. Furthermore, this high density chip should also allow going beyond the classical hypothesis of additivity between genetical and environmental effects on traits.

Thus, the aim of the present study is first to detect QTL influencing egg quality by using this high density chip and, secondly, to observe if the QTL detected could be different according to various environments. To this end, the layers were divided into 2 groups which were fed with two different diets.

### Material and Methods

**Animals and traits.** The population studied consists of 438 sires of a pure line, created and selected by the NOVOGEN breeding company (Le Foeil, France) and of 31,381 of their F1 crossbred female offspring. Hens were hatched in 3 batches in November 2010, May 2011 and November 2011. At 18 weeks of age, they were housed in a production farm in collective cages of 12

half-sibs of sire. They laid from 18 to 75 weeks of age. Half of them were fed ad libitum with a “high energy diet (HE)” with 2881kcal of metabolizable energy (ME) (1342 cages) and the others with a “low energy diet (LE)” with 2455kcal of ME (1346 cages).

Traits were measured by the Zootests company (Ploufragan, France) and concerned 9 variables : Egg Shell Color (ESC), Egg Shell Redness (ESR), Egg Shell Yellowness (ESY), Egg Shell Lightness (ESL), Egg Shell Strength (ESSE), Egg Shell Stiffness (ESSI), Egg Shell Shape (ESSA), Short Length of Egg (SLE) and Egg Weight (EW). They were recorded at 50 and at 70 weeks of age, respectively on 27 747 and on 25 964 eggs (Table 1).

**DNA extraction and genotyping.** Blood samples were collected from brachial veins of sires and DNA was extracted. DNA was hybridized on the 600K Affymetrix® Axiom® HD genotyping array (Kranis et al. (2013)) by Ark-Genomics (Edinburg, UK). In total 438 sires were genotyped for 580,961 markers. These markers covered chromosomes from 1 to 28, two linkage groups (LGE22C19W28\_E50C23 and LGE64), two sex chromosomes and a group of markers with unknown locations. Genotypes were filtered according to 5 successive steps: SNP with a call rate less than 5% were discarded; animals with a call rate less than 95% were excluded; SNP with a MAF less than 0.05 were excluded; SNP with a call rate less than 95% were discarded; SNP significantly ( $P < 5\%$ ) deviated from Hardy-Weinberg Equilibrium were excluded. Thus, 284,643 SNP were kept for the study and no individual was excluded.

**Model.** In a first step, egg measurements were adjusted for environmental effects. Independently for each of the 3 hatches, the estimates of covariables and fixed effects were calculated with the SAS GLM procedure according to the following linear model:

$$Y = \text{Battery} + \text{Column} + \text{Floor} + \text{Experimenter} + \text{Waiting} + \text{Age} + \text{Sire} + E$$

where Y is the phenotypic value, i.e. 9 variables per age; Battery (4 levels), Column (2 levels) and Floor (2 levels) represent the location of the cage in the building; Experimenter (8 to 10 levels) is the person who made the measurement; Waiting and Age are covariables to take into account the waiting time between sample and measurement of egg and the real age of hens (in days); Sire is the fixed effect of the sire (438 levels) and E is the residual. In this model, the diet effect is mingled with the Battery effect.

Raw data were then adjusted for all effects exceeding the significant level of  $P < 0.2$ , except the Sire

effect. Distributions of adjusted data were tested for each trait and extreme individual values, i.e. values that lie at more than 4 phenotypic standard deviations of the mean, were discarded. Finally, for each of the 9 variables, 2 sire performances were calculated as the 2 means of his daughters' performances for each diet separately (HE and LE).

**GWAS analyses.** For each SNP<sub>i</sub> (i=1 to 284,643) and each trait j (j=1 to 18, i.e. 9 traits \* 2 diets), a mixed model taking into account pedigree kinship was applied according to:

$$z_j = \mathbf{1}\mu_{ij} + \mathbf{X}_i\beta_{ij} + e_j \text{ with } V(e_j) = \mathbf{A}\sigma_{uj}^2 + \mathbf{I}\sigma_{ej}^2$$

where  $z_j$  is the vector of the sires performances for the trait j;  $\mu_{ij}$  is the general mean;  $\mathbf{X}_i$  is the incidence matrix of SNP<sub>i</sub> genotypes;  $\beta_{ij}$  is the effect of the SNP<sub>i</sub> on the trait j;  $e_j$  is the residual, a random variable with  $V(e_j)$  variance-covariance matrix;  $\mathbf{A}$  is the pedigree kinship matrix;  $\mathbf{I}$  is the identity matrix;  $\sigma_{uj}^2$  is the genetic variance of the trait j and  $\sigma_{ej}^2$  is the residual variance.

This model was fitted using the BLUPF90 program (Misztal et al. (2002)) and the chromosome wise thresholds ( $P < 1\%$ ) were estimated with the method described by Müller et al. (2011), using the Muller software (Ricard et al. (2013)). Genome wise thresholds ( $P < 5\%$ ) were then calculated according to Bonferroni correction as chromosome wise thresholds at  $P < 0.0015$  (0.05/32 chromosomes).

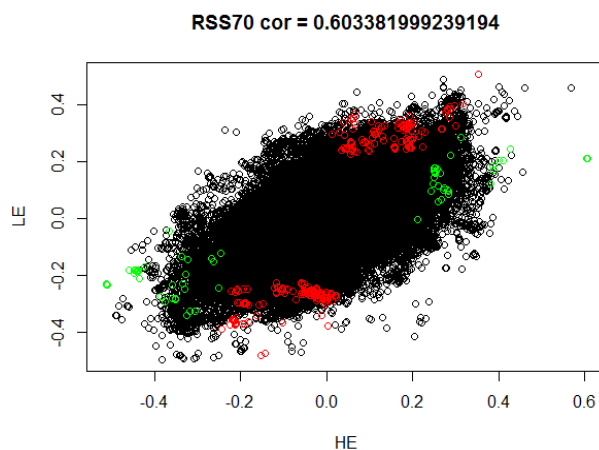
## Results and Discussion

Table 1 shows the mean and the standard deviation for each trait and each diet. No significant differences of mean or of standard deviation between diets were detected.

**Table 1. Number of observations, mean and standard deviation of traits measured at 50 and 70 weeks of age.**

Trait	Number eggs		Mean		SD		
	Diet	LE	HE	LE	HE	LE	HE
ESC50		13580	13528	25.3	24.8	9.5	9.1
ESR50		13605	13546	13.4	13.7	3.0	2.9
ESY50		13493	13445	28.2	28.3	2.6	2.5
ESL50		13596	13548	66.9	66.8	4.6	4.4
ESSI50		12418	12388	191.5	193.4	28.1	29.1
ESSE50		12410	12379	39.6	39.1	7.2	7.4
ESSA50		12403	12375	1.1	1.1	0.02	0.01
SLE50, mm		12409	12378	43.3	43.4	1.3	1.3
EW50, g		13603	13547	60.6	61.1	4.7	4.7
ESC70		12259	12929	25.5	25.4	10.1	9.7
ESR70		12274	12942	13.3	13.4	3.2	3.0
ESY70		12188	12833	28.2	28.3	2.8	2.6
ESL70		12272	12937	67.0	67.1	4.8	4.7
ESSI70		11004	11332	190.5	192.8	29.6	29.2
ESSE70		11007	11327	37.8	37.4	7.5	7.4
ESSA70		10983	11315	1.10	1.10	0.02	0.02
SLE70, mm		11004	11329	43.4	43.4	1.3	1.3
EW70, g		12804	12945	60.6	61.0	4.8	4.9

GWAS analyses showed 9,056 SNP with a chromosome significant ( $P < 1\%$ ) association with at least one egg quality trait. Among these SNP, 1,768 were genome wise significant ( $P < 5\%$ ). Large differences exist between diets and between ages of measurement. Table 2 gives an example of results for ESL. Given a trait, the correlation between estimates of SNP effects, with LE or HE diets, were generally high (up to 0.60) but significantly different of 1. One example is shown in Figure 1 for ESR70. This figure also shows some SNP which have a strong effect ( $P < 1\%$ ) only for one diet.



**Figure 1. Correlation between estimates of SNP effects on Egg Shell Redness at 70 weeks of age (ESR70) with HE diet (in abscissa) and LE diet (in ordinate).**

Chromosome wide significant SNP ( $P < 1\%$ ) with high energy diet are spotted in green and with low energy diet are spotted in red.

### Egg Shell Color (ESC, ESR, ESY and ESL).

For ESC, ESR, ESY and ESL, 2,743 SNP with a significant association were found. On GGA3, 274 SNP in the QTL region detected by Wardecka et al. (2002) have significant association with ESR50 (none with ESR70). At the genome wise level, 72 SNP were found on GGA12, for ESR50, ESC50, and ESL50 for both diet and for ESR70 with low energy diet only. These significant SNP seemed to be split into two different QTL. The first was in a region of 0.623 Mb and the second in a region of 0.289 Mb. The intermediate region was of 7.48 Mb. On GGA10, 161 SNP for ESR50, ESY50, ESL50, ESC50 with high energy diet, ESY50 and ESY70 with low energy diet, were detected at the genome-wise level 5%.

**Table 2. Number of SNP with significant effect ( $P < 1\%$ ) on Egg Shell Lightness (ESL), at 50 or 70 weeks of age and with HE or LE diets, per chromosome.**

Chromo	ESL50	ESL50	ESL70	ESL70
	LE	HE	LE	HE
1	21	2		
2	5		1	
3	3	100		3
4			8	2
5	19	31	25	6
6				
7		6		
8		3	12	1

9	64	45	56	
10	5	168		
11	6	12	12	6
12	106	58	1	10
13		14	5	
14		17	8	
15	1	1	2	
16				
17	4			
18	4	1	28	7
19	19		29	8
20				
21				
22	1	5		1
23	8		70	
24			2	
25	1			
26			1	
27				2
28				
LGE22	10	6		9
LGE64				
Z	59	87	68	3
Unknown	8	7	4	1
Total	344	563	332	59

**Egg Shell Stiffness (ESSI) and Strength (ESSE).** For ESSI and ESSE, 2,162 SNP with a significant association were found. On GGA4, 18 significant SNP for ESSE with low energy diet only were found. Moreover, 119 genome-wise significant SNP for ESSI at 70 weeks of age were detected for low energy diet on GGA1 within 2.85 Mb, a result consistent with the two QTL already found on GGA1 by Tuiskula-Haavisto et al. (2011).

**Egg Shell Shape (ESSA) and Short Length of Egg (SLE).** For ESSA and SLE, 1,803 SNP with a significant association were found. On GGA26, 14 significant SNP for ESSA with high energy diet only were identified. Moreover, 14 genome-wise significant SNP for SLE50 were detected for low energy diet on GGAZ within 0.114 Mb.

**Egg Weight (EW).** For EW, 1,265 SNP with a significant association were found. At 50 weeks of age, 956 SNP were detected and only 417 at 70 weeks of age. For example, on GGA22, 58 SNP with significant association with EW50 were found and none with EW70. Moreover, 17 genome-wise significant SNP for EW50 were identified on GGA11 within 3.677 Mb.

### Conclusion

New QTL for egg quality traits were detected, such as a QTL on GGA12 influencing ESR50, ESY50, ESL50, ESC50 with high energy diet, ESY50 and ESY70

with low energy diet. Although no differences were observed on average level of performances according to the diet, interactions between QTL and diet were observed. All these results should be useful to improve genomic evaluation procedures in layers.

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### Literature Cited

- Chicken QTL database, <http://www.animalgenome.org/cgi-bin/QTLdb/GG/index>.
- Kranis, A., Gheyas, A.A., Boschiero, C., et al. (2013). BMC Genomics, 14:59.
- Misztal, I., Tsuruta, S., Strabel, T., et al. (2002). Proc 7th WCGALP, CD-ROM communication No 28-07.
- Müller, B.U., Stich, B., and Piepho, H.-P. (2011). Heredity, 106:825-831.
- Ricard, A., Filangi, O., and Elsen, J.M. (2013). Muller. Version 3.0.0.
- Tuiskula-Haavisto, M., Honkatukia, M., Preisinger, R., et al. (2011). Anim. Genet., 42:293-299.
- Wardecka, B., Olszewski, R., Jaszczak, K., et al. (2002). J. Appl. Genet., 43:319-329.