



Mapping quantitative trait loci in *Gallus gallus* using principal components

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ABSTRACT - This study aimed at mapping QTL (quantitative trait loci) using linear combinations of characteristics of economical interest in *Gallus gallus*. A total of 350 F₂ chickens from an initial crossing among males from a broiler line (TT) with females from a layer line (CC) were used. It was conducted a QTL mapping in chromosomes of *Gallus gallus* (GGA1, GGA3, GGA5, GGA8, GGA11, and GGA13) for 20 performance and carcass traits. For detecting QTL, it was used the likelihood ratio test between a reduced model (including fixed effects of sex, hatch and random effect of infinitesimal genetic value) and a full model (including all the previous effects plus QTL effects). When original characteristics were analyzed, that is, before the formation of linear combinations, six significant QTLs were mapped at 1% in the genome, four in the GGA1 (live weight at 35 days of age and at 42 days of age, abdominal fat and heart weight); and two on GGA3 (live weight at 35 and 42 days of age); three significant QTLs at 5% in the genome, one on GGA1 (head weight), one on GGA3 (wings weight), and one on GGA8 (gizzard weight); besides seven suggestive linkages for several traits. When QTLs were mapped for principal components, many mapped QTLs were confirmed for original traits, in addition to finding three QTLs and eight suggestive linkages not mapped for the original traits.

Key Words: broilers, canonical variables, chickens, genome

Mapeamento de locos de características quantitativas em *Gallus gallus* com utilização de componentes principais

RESUMO - Este estudo teve por objetivo mapear QTL (*quantitative trait loci*) utilizando combinações lineares de características de interesse econômico em *Gallus gallus*. Foram estudadas 350 aves F₂ oriundas de um cruzamento inicial entre machos de uma linhagem de corte (TT) e fêmeas de uma linhagem de postura (CC). Foi conduzido mapeamento de QTL nos cromossomos *Gallus gallus* (GGA1, GGA3, GGA5, GGA8, GGA11 e GGA13), para 20 características de desempenho e de carcaça. Para detectar QTL foi utilizado o teste da razão de verossimilhanças entre um modelo reduzido (incluindo efeitos fixos de sexo, incubação e o efeito aleatório de valor genético infinitesimal) e um completo (incluindo todos os efeitos anteriores mais os efeitos de QTL). Ao analisar as características originais, ou seja, antes da formação das combinações lineares, foram mapeados seis QTLs significativos a 1% no genoma, quatro no GGA1 (peso vivo aos 35 dias, peso vivo aos 42 dias, gordura abdominal e peso do coração) e dois no GGA3 (peso vivo aos 35 e aos 42 dias de idade); três QTLs significativos a 5% no genoma, sendo um no GGA1 (peso da cabeça), um no GGA3 (peso das asas), e um no GGA8 (peso da moela); além de sete ligações sugestivas para diversas características. Ao mapear QTLs para os componentes principais, foram confirmados vários QTLs mapeados para as características originais, além de encontrar três QTLs e oito ligações sugestivas não mapeadas para as características originais.

Palavras-chave: frangos, galinha, genoma, variáveis canônicas

Introduction

Poultry industry has been able to reach high indices of productivity partly because of advances in animal breeding. Maintaining high levels of genetic gain in poultry is a challenge. In order to do so, molecular genetics may be applied to better understand the complex genetic

architecture that controls quantitative traits as described by De Koning (2008).

A Brazilian population of F₂ animals has been repeatedly analyzed in QTL mapping studies in poultry. Some QTLs mapped in this population have been described by Nones et al. (2006), Ambo et al. (2009) and Campos et al. (2009). Procedures seeking to reduce the occurrence of false

positives or negatives were not employed in these studies, except of course to adjust the level of type I error to an acceptable level.

The literature has suggested using principal components, as described by Mangin et al. (1998), to reduce false negatives (Type II error). This analysis consists in obtaining new characteristics, known as principal components or canonical variables, which are simply linear combinations of the characteristics of interest. According to Mangin et al. (1998), QTL mapping for principal components may find QTLs not mapped when each characteristic is analyzed independently. Therefore, the objective of the present study was to map QTLs for linear combinations of several characteristics of economic interest in poultry in order to independently identify QTLs that are not mapped using characteristics of economical interest.

Material and Methods

The reference population in the present study was developed at Embrapa Suínos e Aves in Concórdia, SC, Brazil using the line-cross model described by Knott & Haley (1992). A broiler line (TT) was selected for six generations for body weight, feed conversion, feed consumption, yields of carcass and parts, viability, fertility, eclodibility, reduced body fat and reduced occurrence of metabolic diseases. A layer line (CC) was selected for eight generations for egg production, egg weight, feed conversion, viability, sexual maturity, fertility, eclodibility, egg quality and reduced body weight. The F₂ chicken population used in this study was developed by crossing

the broiler line (TT) with the layer line (CC). Four F₂ families (350 chickens) were selected for genotyping chromosomes 1, 3, 5, 8, 11 and 13 using microsatellite markers.

The F₂ population was raised as broilers, and received feed and water *ad libitum*. Animals received a starter ration from 1 to 21 days of age (21% crude protein and 3,150 Kcal/kg metabolizable energy), growth ration from 22 to 35 days of age (20% crude protein and 3,200 Kcal/kg ME), and a finishing ration from 36 to 41 days of age (18.5% crude protein and 3,200 Kcal/kg ME). Chicks were kept in collective boxes until 34 days of age. From the 35th day until the 41st day, animals were kept in individual cages to evaluate individual feed intake.

Linkage maps (Table 1) were obtained using CRIMAP software according to Green et al. (1990).

Live weight at 1, 35 and 42 (LW1, LW35, and LW42) days of age, weight gain (WG35/41), feed intake (FI35/41) and feed conversion (FC35/41) from day 35 to 41 were performance traits evaluated. Body weight at 42 days of age was measured after 6 hours of fasting and transportation to slaughter house. Carcass traits evaluated after four hours of refrigeration consisted of weight (g) of lungs, liver, heart, gizzard, breast, legs (weight of thigh and drumstick), carcass (without viscera, feet and head), residual carcass (weight of carcass without breast, wings and legs), wings, head, feet and abdominal fat and intestinal length (cm). Hematocrit values were determined in the laboratory using blood collected at slaughter.

To test sex effects, hatch, family and the covariable LW42, an analysis of variance was used, in which the residuals were initially evaluated assuming normal distribution,

Table 1 - Position of microsatellite markers in their respective chromosomes

GGA	Marker	cM	GGA	Marker	cM	GGA	Marker	cM
1	MCW0010	0	1	LEI0106	315.0	5	MCW0193	19.9
1	MCW0208	3.4	1	ADL0183	328.0	5	MCW0090	28.5
1	ADL0188	61.3	1	LEI0079	332.0	5	LEI0145	64.2
1	ADL0234	73.7	1	MCW0145	340.0	5	LEI0149	84.2
1	LEI0068	79.6	1	MCW0020	343.0	5	ADL0233	97.4
1	MCW0297	85.8	1	ROS0025	392.1	5	ADL0298	151
1	LEI0146	90.3	3	LEI0043	0	8	ABR0322	0
1	LEI0174	120.0	3	MCW0169	20	8	MCW0095	18.2
1	MCW0018	121.0	3	MCW0222	84	8	ADL0154	34.1
1	ADL0150	124.0	3	LEI0161	108	8	ABR0345	41.6
1	ADL0319	131.0	3	LEI0029	111	8	ADL0172	82.6
1	MCW0058	155.0	3	ADL0371	135	11	LEI0143	0
1	LEI0071	156.0	3	LEI0118	163	11	ADL0123	23.5
1	LEI0138	170.0	3	MCW0277	166	11	ADL0210	62.9
1	MCW0068	191.0	3	ADL0127	168	11	MCW0230	106
1	ADL0020	196.0	3	MCW0224	198	13	ADL0147	0
1	LEI0160	206.0	3	MCW0207	217	13	MCW0213	10.5
1	ADL0148	232.0	3	MCW0040	246	13	LEI0251	17.2
1	MCW0036	261.0	3	LEI0166	273	13	MCW0110	34
1	LEI0169	280.0	5	LEI0082	0	13	MCW0104	57

GGA (*Gallus gallus* chromosome).

homogeneous variance and outliers using SAS Guided Data Analysis (SAS, 2002). Means were adjusted using the following model:

$$Y_{ijk} = S_i + H_j + F_k + c_{ijk}LW42 + e_{ijk}$$

in which Y_{ijk} is the phenotypic value of a characteristic of the ijk^{th} individual, S_i is the fixed effect of the i^{th} sex, H_j is the fixed effect of the j^{th} hatch, F_k is the fixed effect of the k^{th} family of full sibs, c_{ijk} is the coefficient corresponding to the effect of the covariable LW42 of the ijk^{th} individual and e_{ijk} is the error, NID $(0, \sigma_e^2)$. The different factors were evaluated using GLM procedure (SAS, 2002). The covariable LW42 was not included in the model for weights at 1, 35 and 42 days of age. Means, standard deviations, coefficients of variance, as well as maximal and minimal values were obtained using PROC MEANS (SAS, 2002).

Sequences of principle components analyses were carried out to separate the 20 traits into four groups. In this way, a characteristic was removed after each analysis in order to maximize the variance explained by the first principle component. It was established that each group would have a minimum of four traits, since simultaneously mapping multiple traits works reasonably well with up to three traits and the results are easier to interpret (Gilbert and Le Roy, 2003). Group 1 (G1) was formed by breast, thigh, wings, carcass and residual carcass weights as well as LW42 and LW35. Group 2 (G2) included head, feet, liver, heart and gizzard weights. Group three (G3) consisted of lungs weight, abdominal fat as well as weight gain and feed intake from 35 to 41 days of age. Lastly, group 4 (G4) included weight at 1 day of age, intestinal length, hematocrit values and feed conversion from 35 to 41 days of age.

Canonical variables were obtained from the original traits using PROC PRINCOMP (SAS, 2002). A correlation matrix was used to obtain the eigenvectors, since the original traits sometimes present variances of very different magnitudes. Principle components analysis seeks to obtain canonical variables (principle components) so that the first canonical variable corresponds to the greatest eigenvalue, the second canonical variable to the second greatest and so on (Table 2). Each characteristic used in the principal components analysis has a determined contribution to the canonical variable in question (Table 3). These values will be useful in interpreting the QTL map of the canonical variables, as it will be shown.

For QTL mapping, a likelihood ratio test was performed between two hierarchical models. The complete model is as it follows:

$$Y_{ijk} = \mu + S_i + H_j + a_{ijk}LW42 + b_{ijk}I + c_{ijk}A + d_{ijk}D + e_{ijk}$$

in which Y_{ijk} = the phenotypic value of a characteristic of the ijk^{th} animal, μ = the general mean of the phenotype considered, S_i = the fixed effect of the i^{th} sex, H_j = the fixed effect of the j^{th} hatch, a_{ijk} = the coefficient associated with the covariable LW42 of the ijk^{th} animal, b_{ijk} = the coefficient associated with the random effect of infinitesimal genetic value (I) of the ijk^{th} animal, c_{ijk} = the coefficient associated with the additive effect (A) of the QTL in the ijk^{th} animal, d_{ijk} = the coefficient associated with the dominant effect (D) of the QTL in the ijk^{th} animal and e_{ijk} = the random residual of the model in the ijk^{th} animal. The reduced model is nothing more than the complete model without the parameter of interest under study. It is important to mention that in order to avoid super-parametrization of the model, it was opted for maintaining only the significant effects in the analysis of variance.

The Qxpak software (Perez-Enciso & Misztal, 2004) was used for QTL mapping. Significance levels of mapped QTLs were estimated according to Lander and Kruglyak (1995), and the confidence interval for QTL location was obtained as described by Mangin et al. (1994). Phenotypic variation explained by the QTL was calculated as described by Sorensen et al. (2003).

Table 2 - Eigenvalues and percent variance explained by each principal component and percent accumulated variance

Principal component (PC)	Eigenvalues	% Variance	Accumulated variance
Group 1			
PC1	6.52	93.14	93.14
PC2	0.12	1.69	94.83
PC3	0.11	1.60	96.44
PC4	0.11	1.53	97.97
PC5	0.07	1.00	98.97
PC6	0.05	0.78	99.75
PC7	0.02	0.25	1.00
Group 2			
PC1	3.48	69.53	69.53
PC2	0.66	13.16	82.68
PC3	0.40	8.03	90.71
PC4	0.34	6.78	97.49
PC5	0.13	2.51	100
Group 3			
PC1	2.48	62.12	62.12
PC2	0.66	16.62	78.74
PC3	0.57	14.33	93.07
PC4	0.28	6.93	100
Group 4			
PC1	1.21	30.22	30.22
PC2	1.11	27.85	58.06
PC3	0.93	23.17	81.23
PC4	0.75	18.77	100

Table 3 - Contribution of traits to principal components

Trait	Principal component 1	Principal component 2	Principal component 3	Principal component 4	Principal component 5	Principal component 6	Principal component 7
			Group 1				
Live weight at 42 days of age	15.06	0.01	0.67	0.33	0.17	6.16	77.59
Live weight at 35 days of age	13.93	39.34	33.03	0.72	7.92	2.60	2.47
Wings	14.17	5.75	14.72	23.37	35.30	5.18	1.51
Thighs + Drumsticks	14.32	4.70	9.16	16.05	32.50	16.72	6.55
Breast	14.02	7.54	12.20	53.75	1.01	8.64	2.84
Residual carcass	14.08	29.85	28.08	0.47	15.52	3.64	8.36
Carcass	14.42	12.81	2.13	5.30	7.57	57.07	0.68
			Group 2				
Head	23.31	0.89	5.61	34.26	35.93		
Feet	25.07	0.56	3.36	10.39	60.62		
Liver	20.54	0.74	26.91	50.58	1.23		
Heart	16.34	38.15	43.24	2.25	0.03		
Gizzard	14.74	59.66	20.88	2.53	2.19		
			Group 3				
Abdominal fat	18.90	74.91	5.12	1.07			
Lungs	22.48	0.04	76.52	0.96			
Weight gain from 35 to 41 days of age	31.00	6.10	4.95	57.94			
Feed intake from 35 to 41 days of age	27.62	18.94	13.41	40.03			
			Group 4				
Live weight at 1 day of age	30.26	0.03	62.85	6.85			
Intestine	8.96	50.98	13.14	26.93			
Feed conversion from 35 to 41 days of age	9.53	48.96	15.82	25.69			
Hematocrit	51.25	0.03	8.19	40.53			

Results and Discussion

Traits showed coefficients of variation higher than averages observed by Mohallem et al. (2008) (Table 4), which is due to the elevated effect of sex, family and hatch, but that is mainly due to the fact of dealing with an F2 population originating from crossing phenotypically distinct lines. Differences between minimal and maximal values were

expected and desired, since variability in the F2 population is needed to map QTL.

Average live weight at 42 days of age is lower than the average observed in commercial lines by Havenstein et al. (2003), which was expected from a broiler × layer cross. On the other hand, this value is near that observed by Zerehdaran et al. (2004) in an experimental F₂ population whose founder lines varied for live weight.

Table 4 - Descriptive statistics of the traits under study

Trait	Average	Standard deviation	Coefficient of variation	Minimum	Maximum
Live weight at 1 day of age (g)	44.57	4.10	9.20	32.50	55.50
Live weight at 35 days of age (g)	804.92	136.84	17.00	458.00	1341.00
Live weight at 42 day of age (g)	988.85	182.51	18.46	402.00	1688.00
Weight gain (g)	221.74	66.08	29.80	4.00	418.00
Feed intake (g)	602.36	134.23	22.28	258.00	1176.00
Feed conversion (kg)	2.84	0.69	24.38	1.35	7.55
Weight of head (g)	34.76	5.89	16.94	18.00	55.00
Weight of feet (g)	40.15	8.82	21.96	16.00	74.00
Weight of wings (g)	82.29	14.61	17.76	31.00	140.00
Weight of thigh + drumstick (g)	212.18	43.83	20.66	67.00	354.00
Weight of breast (g)	161.95	34.85	21.52	55.00	303.00
Weight of carcass (g)	643.98	128.09	19.89	234.00	1175.00
Weight of residual carcass (g)	187.26	38.39	20.50	75.00	350.00
Weight of abdominal fat (g)	16.32	7.74	47.44	1.00	55.00
Weight of liver (g)	26.64	5.20	19.53	15.00	49.00
Weight of heart (g)	6.42	1.64	25.57	2.00	12.00
Weight of gizzard (g)	24.32	4.49	18.45	13.00	39.00
Weight of lungs (g)	8.08	2.23	27.62	3.00	16.00
Intestine length (cm)	153.80	14.96	9.73	111.00	204.00
Hematocrit	27.88	2.94	10.54	18.00	42.00

QTLs of small effect, i.e. those that explain reduced percentages of phenotypic variance, are more difficult to map (Gilbert & Le Roy, 2003). In these cases, larger samples, denser genetic maps or alternative QTL mapping methods are needed for increasing power of statistical test. Mangin et al. (1998) demonstrated that using canonical variable is a good approach for increasing the power of detecting QTLs. To verify the effect of this alternative methodology, QTL of original traits, i.e. before obtaining linear combination, were mapped (Table 5) for consequent comparison to principal component analysis results. Percentages of phenotypic variance explained by the QTLs varied from 1.37 to 7.98%. It may be expected that some QTLs may not have been mapped for original traits in the present study (350 animals and medium density of a marker every 17.7 cM) due to the reduced power of the statistical test.

Adjustment of infinitesimal genetic value is extremely important to the present analysis, since non-isolation of the polygenic effect may obtain biased estimates of the QTL effects. The infinitesimal effect was not adjusted in works by Nones et al. (2006), Ambo et al. (2009) and Campos et al. (2009) which may have led these studies to detect false positive QTLs or to overestimate the effects of QTLs. For example, Campos et al. (2009) identified QTL for abdominal fat at 115cM on chromosome 3, explaining 4.08% phenotypic variance of this trait with significant additive, dominance and imprinting effects. This may be the QTL that was detected in the present study at 115 cM, although with a significant additive effect and explaining approximately 2.33% phenotypic variance. These differences may result from adjustment in the infinitesimal genetic effect used in the present study and not considered in the work by Campos et al. (2009).

It is important to stress that canonical variables were used to identify new QTLs that had not been mapped in analysis of the original traits, i.e. before obtaining linear combinations. Consequently, interest of the present study is restricted to increasing the power of the test as described in Weller et al. (1996), Magin et al. (1998) and Gilbert & Le Roy (2003), and not in any specific linear combination. Two highly significant linkages, four significant linkages and ten suggestive linkages were mapped for canonical variables (Table 6). Since the criteria for suggestive linkage leads to one false positive throughout the genome (Lander & Kruglyak, 1995), it is possible to conclude that at least nine of these suggestive linkages are real QTLs. Concerning principal components, only QTL positions were described because both dominance and imprinting effects were not significant, while additive effects are associated with linear combinations, but no with original traits.

Four QTLs and one suggestive linkage were mapped for Group 1 canonical variables. The first QTL is located at 79 cM of GGA1 and it is associated with PC1 (Table 6). Apparently, this QTL has an effect on all traits used in forming this component since contributions of seven original traits for PC1 are very similar (Table 3). However, QTL results from original traits (Table 5) show that QTLs in this region of GGA1 only exist for LW35 and LW42. Based on the considerable fall in nominal probability, evidenced by the significance level of the test for LW35 and LW42 versus the test for PC1 in GGA1, it is possible that the QTL mapped here is the same one found in analyses of LW42 and LW35, and that the other traits included in the linear combination reduced the nominal probability.

Table 5 - QTLs mapped for performance and carcass traits

Trait	GGA ¹	(Position ²) confidence interval	Additive effect	Standard error	VP% ³
Weight of head	1	105 (162*) 190	-0.60	0.16	2.93
Weight of feet	1	174 (197 [†]) 210	-0.56	0.16	1.37
Weight of heart	1	68 (74**) 77	0.19	0.07	1.67
Weight of abdominal fat	1	89 (91**) 101	-1.39	0.28	3.50
Weight gain	1	1 (15 [†]) 39	-10.54	2.90	3.69
Live weight at 42 days of age	1	65 (81**) 111	32.41	8.55	2.64
Live weight at 35 days of age	1	67 (81**) 103	28.72	6.67	3.49
Feed conversion	1	1 (15 [†]) 37	0.16	0.04	3.24
Weight of wings	3	138 (151*)166	-1.25	0.31	3.96
Weight of thigh + drumstick	3	124(146 [†])167	-2.94	0.92	3.27
Weight of abdominal fat	3	95 (115 [†]) 133	1.14	0.34	2.33
Live weight at 42 days	3	50 (98**) 118	49.13	11.12	6.03
Live weight at 35 days	3	87 (98**) 113	43.47	8.66	7.98
Weight of gizzard	8	37 (58*)75	-1.14	0.30	6.79
Weight of wings	11	59 (96 [†])105.5	1.62	0.42	5.83
Weight of heart	13	46 (57 [†])57	0.32	0.09	4.54

¹ *Gallus gallus* chromosome.

² ** 1% genomewide, * 5% genomewide and [†] suggestive linkage.

³ VP% – percent phenotypic variance explained by QTL.

Table 6 - QTL with significant or suggestive effects on canonical variables

Principal component (PC)	Group	GGA ¹	(Position ²) & confidence interval
PC1	1	1	63 (79*) 112
PC4	1	1	356 (376*) 393
PC1	2	1	66 (83**) 99
PC2	2	1	100 (117 [†]) 165
PC3	3	1	88 (91 [†]) 98
PC4	3	1	166 (180 [†]) 199
PC1	4	1	2 (32 [†]) 51
PC2	4	1	332 (340 [†]) 366
PC1	1	3	44 (62**) 114
PC3	1	3	94 (126*) 159
PC4	1	3	136 (152 [†]) 166
PC1	2	3	62 (111*) 123
PC1	3	3	146 (166 [†]) 190
PC2	3	3	37 (112 [†]) 124
PC4	3	3	91 (110 [†]) 124
PC2	4	11	12 (36 [†]) 58

¹ GGA – *Gallus gallus* chromosome.

² **1% genomewide. *5% genomewide and [†]suggestive linkage.

The second QTL for Group 1 was mapped for 376 cM of GGA1 and it is associated with PC4 (Table 6). It is a novel QTL since this region of GGA1 did not have any suggestive linkages in the analysis of the original traits composing Group 1. This principal component greatly contributes to the weight of wings, thighs and drumsticks and breast (Table 3). However, this region of GGA1 should be refined to identify and to explain the effect of this QTL and the characteristic(s) which it affects.

The third QTL for G1 was mapped at 62 cM of GGA3 and it is associated with PC1. Its effect was highly significant and near the region where other highly significant QTLs for live weight at 35 and 42 days of age have been mapped (Table 5). Since the probability of the nominal test was not altered, it is possible that this QTL has a pleiotropic effect on some traits of this principal component or even on all of them. The fourth QTL of G1 was mapped at 126 cM of GGA3 and it is associated with PC3. The power of the test can be seen now since this region of GGA3 did not have QTL for the traits that compose G1. This principal component received great contribution from the residual carcass weight and LW35 (Table 3). Consequently, inclusion of more markers in this region of GGA3 may help identify the traits influenced by this QTL.

Suggestive linkage for canonical variables of G1 was mapped at 152 cM from GGA3 and it is associated with PC4. Wings, thighs and breast are traits with important contribution for PC4 (Table 3). Since a QTL was mapped on GGA3 for wings at 151 cM and suggestive linkage for thighs at 146 cM (Table 5), this suggestive linkage at 152 cM for PC4 confirmed results found with original trait.

Two QTLs and a suggestive linkage were mapped for G2 (Table 6). The first QTL is located at 83 cM of GGA1 and it is associated with PC1. This component received great contribution from head, feet, liver, heart and gizzard weights (Table 3). Furthermore, a QTL for heart weight was mapped at this region at 74cM (Table 5). Evidence from Navarro et al. (2005) indicates QTL for heart weight near this region. Since there was no reduction in nominal probability, it is possible that this QTL also has an effect on some other trait(s) that contribute to the variation explained by this principal component. The second QTL influencing traits of G2 is located at 111 cM of GGA3 and also has an effect on PC1. It is a new QTL, since no QTL were mapped in GGA3 in the analysis of the original traits composing G2. However, it is possible that this QTL has a pleiotropic effect on some traits in this group. Suggestive linkage with some influence on G2 was mapped at 117cM from GGA1 and it is associated with PC2 (Table 6). This component was mainly influenced by heart and gizzard weights (Table 3) which did not have any QTL in this region of GGA1. It is possible that this QTL has a pleiotropic effect on these two traits, although Navarro et al. (2005) mapped QTL only for gizzard weight in this same region. Consequently, inclusion of more markers in this region of GGA1 or increasing the F2 population may help make the effects of this QTL clear.

Five suggestive linkages for the G3 canonical variable were mapped (Table 6). The first for PC3 at 91cM of GGA1 confirms the QTL mapped at 91 cM for weight of abdominal fat (Table 5). This component is strongly influenced by variation in lungs weight and to a lesser degree by variations in weight of abdominal fat, weight gain and feed intake (Table 3). The contribution of lungs weight to PC2 can explain the reduced nominal probability, which stopped being highly significant in QTL analysis of weight of abdominal fat to become suggestive in analysis of PC3 in G3. The second suggestive linkage is located at 180 cM of GGA1 and has an effect on PC4. Not even suggestive linkages for original traits composing G3 have been found in this region of GGA1. This suggestive linkage may possibly affect weight gain and/or feed intake. Both of which greatly contribute to the amount of variation PC4 explains in G3 (Table 3). Therefore, refining this GGA3 region may help identify and clarify the effects of this suggestive linkage. The third suggestive linkage of G3 is associated with PC2 and located at 112 cM of GGA3. This linkage may result from the effects of suggestive linkage mapped at 115 cM of GGA3 for abdominal fat (Table 5), since variation in abdominal fat has important contribution to PC2 in G3 (Table 3). The fourth suggestive linkage was mapped at 166 cM of GGA3 and it is associated with PC1. This principal component

received great contribution from the four traits forming this group (Table 3). When evaluated singularly, no QTL were mapped for any of the four traits in the group. Thus, the linkage mapped may have an effect on any of the traits in G3 or even a pleiotropic effect on some of them. The last suggestive linkage for G3 was mapped at 110 cM of GGA3 and it is associated with PC4, which received strong contribution from weight gain and feed intake. When these traits were analyzed individually, no QTL were mapped. More studies of this region of GGA3 are needed since feed intake and weight gain are traits of economic interest.

Three suggestive linkages were mapped for G4 canonical variables (Table 6). The first is located at 32 cM of GGA1 and has an effect on PC1. This suggestive linkage receives an important contribution from hematocrit and weight at 1 day (Table 3). However, no QTL were mapped for these traits, when analyzed individually (Table 5). The remaining suggestive linkages for G4 were observed for PC2 at 340 cM of GGA1 and 36 cM of GGA11. These principle components received great contribution from variations in intestine length and feed conversion (Table 3). Both linkages may be false positives or novel QTLs found through linear combinations since no QTLs or suggestive linkages for G4 traits were mapped for these regions of GGA1 and GGA11. Consequently, these regions should be studied further.

Interpretation of QTL analysis results for canonical variables is difficult when the original variables are interesting for studying and not the linear combinations. For example, the QTL for PC1 in G1 at 79cM of GGA1 had a negative additive effect (data not shown). Analysis and comparison of QTL analysis results of the original traits leads to the conclusion that this QTL is the same as that mapped with a positive additive effect for LW35 and LW42. However, the effects of QTLs associated with linear combinations do not have an immediate interpretation. The same problem is observed for the percent of variance explained by the QTL. Thus, in cases in which the linear combination is not very important, QTL results for canonical variables may only be used to identify regions with potential QTLs. In the present study, use of canonical variables allowed the identification of three QTLs not mapped with the original traits and eight new suggestive linkages. These regions may now be refined with a greater number of molecular markers in the expectation of obtaining more information about the effect of these QTLs on the original traits. However, the use of linear combinations will not necessarily increase the number of QTLs or the suggestive linkages found and it may even reduce this number. For

example, the suggestive linkages for wings and heart weights respectively located at 96 cM of GGA11 and 57 cM of GGA13 were not identified in the principal components analysis. Consequently, the linear combinations should be carefully defined to maximize the capacity of locating QTLs.

Conclusions

Group composition, for principal component analysis, should be carefully planned, to avoid reduction of the power of the test in the mapping analysis.

All significant effects found for canonical variables must be compared to those found when working with the original traits.

Mapping loci of canonical variables helps identify significant regions that are not mapped in isolated analysis of the original traits.

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