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The Combine Effect of Mutation in Lipoprotein Lipase and ApoVLDL-II Genes on Meat Quality

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Abstract: Meat quality such as tenderness, color Density (OD), pH and Water Holding Capacity (WHC) were estimated from breast muscle of genetically fat and lean chickens at 12 week of age. Mutation in lipoprotein lipase and apoVLDL-II genes was detected by PCR-SSCP techniques. Agreement of the genotype frequencies with the Hardy-Weinberg equilibrium expectations was tested using a chi-square goodness of -fit test. Lipoprotein lipase gene frequency was significantly different ($p<0.01$) in Rugao population, whereas in apoVLDL-II gene there are no significantly different between populations. The populations were differed significantly ($p<0.01$) within two genes. Lipoprotein lipase genotype was significantly ($p<0.05$) effect water holding capacity and meat tenderness. However, apoVLDL-II genotype was non significantly affected meat quality traits. The results also indicated that the interaction of Lipoprotein lipase and apoVLDL-II genotype was significantly ($p<0.01$) effected color density, pH and meat tenderness, whereas it was non significantly effects water holding capacity of breast muscle.

Key words: Lipoprotein lipase, apoVLDL-II, meat quality, genotype, mutation

INTRODUCTION

Growth and meat yield traits in poultry, as well as fitness traits, are controlled by many genes Quantitative Trait Loci (QTL). The total effect of the QTLs is influenced by many genes that might interact with each other and the environment that might interact with the genotype (Cahaner, 1990). The possibility of genetically improving carcass quality by selection depends on the genetic variability of body weight and body composition. Meat quality is usually determined in breast muscles are crucial for the culinary value and technological properties of chicken meat and have been investigated by many authors (Knust and Pingel, 1992).The physiological actions of LPL in catabolism of Chylomicrons (CMs) and VLDL and in the production of much of the lipids and apolipoproteins that form HDL have been appreciated for more than a decadev (Goldberg, 1996). VLDL and HDL concentration are higher in the fat birds whether fed or starved. High plasma levels of VLDL or triglycerides are frequently associated with genetic or nutritional factors that induce excessive hepatic lipogenesis (Leclercq, 1984). Thus, LPL inhibition results in a decrease in the esterified cholesterol percentage and an increase in triglycerides. Although fatty liver is associated with reduced egg production and increased mortality in laying hens (Hermier, 1997). Adipose tissue growth in birds depends mainly on the availability of triglycerides transported by VLDL. This may be the result of increased adipose triglyceride synthesis, because adipose tissue in LPL-deficient humans contains less essential fatty acid (e.g., linoleic acid) (18:2) than that of normal persons (Ullrich *et al*., 2001). This was confirmed in a mouse model with adipose LPL deficiency. Because both humans and mice without adipose tissue LPL still have some essential fatty acid in the adipose tissue, there must be other pathways mediating organ fatty acid uptake (Jaccoby *at el*., 1996). The objective of this experiment was to investigate the interaction effect of lipoprotein lipase and apoVLDL-II genes on meat quality.

MATERIALS AND METHODS

Animals and genetic analysis: A total of 120 chickens from Anka and Rugao breed were taken as a representative sample, at Jiangsu Poultry Institute, Yangzhou, China. Carcasses were dissected manually therefore; Water Holding Capacity (WHC), tenderness, color Density (OD) and pH were estimated from breast muscle (Musa *et al*., 2006). DNA was isolated from the whole blood according to the methods described by (Sambrook *et al*., 1989). Primers for a candidate apoVLDL-II and Lipoprotein lipase genes were design using Oligo 6.0 software Table 1, PCR-SSCP was developed for mutation detection.

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Table 1: Primers sequences, location, product size and annealing temperature of candidates (LPL and apoVLDL-II) genes

Gene	Sequences (5-3 flanking region)	location	Product size	Annealing temperature
LPL	F GCTGAGTTTTCTTGGGAGTTGGG R GCCTTGCTCCCTTGAATGTTTG	17822-15217	395bp	59.8°C
ApoVLDL-II	F ATTGACTAGCGTGAGATTCC R ATGATGGTGCAGTTCTTCTT	2788-3071	303 bp	57°C

F and R refers to forward and reverse primers

Statistical analysis: Genotype and gene frequency were estimated according to Cerit *et al*. (2004). Agreement of the genotype frequencies with the Hardy-Weinberg equilibrium expectations was tested using a chi square goodness of -fit test using Chi-Square calculator V 1.51. The associations of meat quality traits with apoVLDL-II and lipoprotein lipase genotype were determined following this model Yij = μ+ Mi + eij, using General Linear Model of SAS 9.0 software. The following model was also fitted to study the interaction effect of apoVLDL-II and lipoprotein lipase genes genotype with meat quality. Yij = μ + Mi + breed + LPL + ApVLDL-II + [LPL x apoVLDL-II] + eij where Yij is phenotypic value of (meat quality), μ is population mean, Mi is the fixed effect of the ith genotype and eij is random error effect of each observation, it was determined by ANOVA using General Linear Model GLM, all analysis was performed by SAS 9.0 software.

RESULTS AND DISCUSSION

PCR-SSCP genotype: PCR-SSCP analysis of candidate Lipoprotein lipase and apoVLDL-II genes were presented in Fig. 1 and 2, respectively. Their genotype and gene frequency were similarly estimated Table 2. Genotype frequency of lipoprotein lipase was in agreement with Hardy-Weinberg equilibrium expectations in Rugao population, whereas in apoVLDL-II gene it was non significantly different between populations. It may be due to the phenotypic difference of breeds. However there are no significantly different between populations in apoVLDL-II gene. Breeds were differed significantly ($p<0.01$) in lipoprotein lipase and apoVLDL-II genes. The multiple genes have been identified as a candidate can be used to improve animal traits through Markers Assisted Selection (MAS) on genotype (Dekkrs, 2004).

The interaction effect of LPL and apoVLDL-II genes on meat quality: Lipoprotein lipase genotype was significantly ($p<0.05$) effect water holding capacity and meat tenderness. However, apoVLDL-II genotype was non significantly effected meat quality traits Table 3. The interaction of Lipoprotein lipase and apoVLDL-II genotype effect on meat quality was investigated and deposit in Table 4. The result observed that an individual with heterozygous LPL and apoVLDL-II genotype has

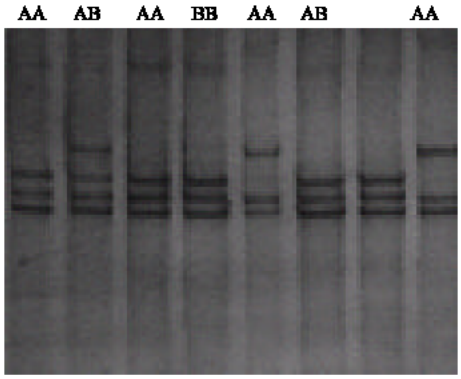

Fig. 1: PCR-SSCP of LPL primer

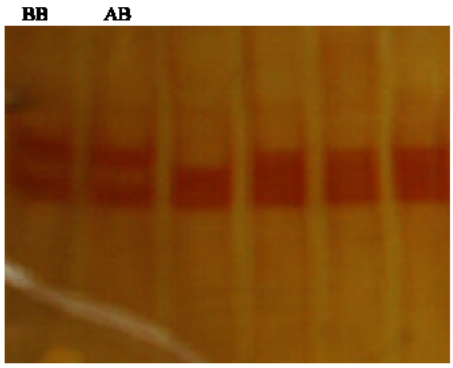

Fig. 2: PCR-SSCP of apoVLDL-II primer

significantly high OD compared with those homozygous LPL and homozygous or heterozygous apoVLDL-II. The pH of meat quality was significantly ($p<0.05$) lower in individual with homozygous LPL and apoVLDL-II compared with those are homozygous LPL and heterozygous apoVLDL-II. There are no significant effects of the interaction of genes on water holding capacity of breast muscle. In addition tenderness was significantly affected by the interaction of genes. In our previous study we show that breeds were differed significantly ($p<0.05$) in color density, pH and tenderness (Musa *et al*., 2006). Color and water holding capacity influence perceptions of the acceptability of the meat product but vary with the species, muscle function, age of the animal and storage conditions (Miller, 1994; Berri, 2000). Rudas *et al*. (1972) indicate that obese chicken displayed higher plasma concentrations of glucose, total lipid, cholesterol

Table 2: Genotype and gene frequency of lipoprotein lipase and apoVLDL-II genes in chicken populations

Population	No.	AA	AB	BB	X^2	A	B	X^2
Lipoprotein lipase								
Anka	59	0.254	0.271	0.475	17.09**	0.389	0.611	5.49
Rugao	59	0.627	0.169	0.203		0.712	0.288	9.38**
ApoVLDL-II								
Anka	59	0.644	0.322	0.034	16.58**	0.805	0.195	0.02
Rugao	59	0.271	0.644	0.085		0.593	0.407	3.86

** Chi-square value was significance at ($p<0.01$)

Table 3: Means and standard errors of various genotypes for meat quality

Loci	Genotype	n	OD	pH	WHC	Tenderness
Lipoprotein lipase						
	TT	52	0.715±0.046	5.680±0.013	0.332±0.006a	2.779±0.100a
	TC	26	0.788±0.064	5.714±0.019	0.309±0.009b	3.250±0.141b
	CC	40	0.725±0.052	5.714±0.015	0.321±0.007	3.007±0.114
ApoVLDL-II						
	TT	54	0.796±0.044	5.716±0.013	0.322±0.006	3.107±0.098
	TC	57	0.717±0.046	5.691±0.014	0.321±0.007	2.985±0.103
	CC	7	0.739±0.141	5.690±0.042	0.315±0.020	2.914±0.314

Different superscripts means significant difference ($p<0.05$); n, sample size, OD, color density; WHC, water holding capacity,

Table 4: Means and standard errors of various interaction genotypes for meat quality

Lipoprotein lipase	ApoVLDL-IIn	OD	pH	WHC	Tenderness
TT	TT	200.797±0.072	5.698±0.021	0.340±0.010	2.979±0.159
	TC	280.666±0.061	5.665±0.018a	0.327±0.009	2.696±0.135a
	CC	40.649±0.160	5.695±0.048	0.329±0.023	2.368±0.357ad
TC	TT	140.777±0.086	5.718±0.026	0.309±0.012	3.240±0.191b
	TC	100.878±0.101a	5.711±0.030	0.312±0.014	3.394±0.226c
	CC	20.412±0.227	5.705±0.068	0.297±0.032	2.596±0.504
CC	TT	200.815±0.072ab	5.733±0.021b	0.317±0.010	3.103±0.159
	TC	190.608±0.074c	5.697±0.022	0.326±0.010	2.866±0.164

Different superscripts means significant difference ($p<0.05$); n, sample size, OD, color density; WHC, water holding capacity

and phospholipids. Grey *et al.* (1986) who compared lines of turkeys with different growth rates; he concluded that some of turkeys were observed variations in tenderness might result from genetic differences among birds. However, Latif *et al.* (1998) indicated that it was due to interaction between genotype and rearing condition. Tenderness was observed positively correlated with color density (Barbut, 1996; Li Bihan *et al*., 1998). The lower pH of chicken could be due to the better welfare conditions that reduce the stress pre-slaughter and thus consumption of glycogen (Castellinri *et al*., 2002). It could be supposed that genetic strain has a role in the improvement of customer appraisal of poultry meat (Abenni and Bergrgoglio, 2001). Kirchgessner *et al.* (1992) found the slight improvements of juiciness and overall classification of breast meat with high dietary levels of linoleic acid. Especially tenderness was observed positive correlated with color density (LeBihan-Duval *et al*., 1998).

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