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A Study of Growth Performance, Carcass Characteristic, Meat Quality and Association of Polymorphism in the *ApoVLDL-II* Gene with Fat Accumulation in the Female Broiler, Thai Native and Betong Chickens (KU Line)

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***Abstract*—** Both Betong chicken (KU Line) and Thai Native chickens were the high quality of the meat and low carcass fat compared to broiler chickens. The objective of this study was to determine the growth performance, carcass characteristic, meat quality and association of polymorphism in the *ApoVLDL-II* gene with fat accumulation in the female broiler, Thai Native and Betong (KU line) chickens at 4-14 weeks. The chickens were used and reared under the same environment and management (100 chicks per breed). The results showed that body weight (BW) of broiler chickens was significantly higher than Thai Native and Betong (KU line) chickens ($P < 0.01$) through all the experiment. At 4-8 weeks of age, feed conversion ratio (FCR) of broiler chickens was significantly better than Thai Native and Betong (KU line) chickens ($P < 0.01$), then increased at week 8-14. The percentage of breast, abdominal fat and subcutaneous fat of broiler chickens was significantly greater than Thai Native and Betong (KU line) chickens ($P < 0.01$). However, Thai Native chickens showed the highest percentage of liver ($P < 0.01$) when compared to other breeds. In addition, the percentage of wing of Thai Native and Betong (KU line) chickens were significantly ($P < 0.01$) higher than broiler chickens. Meat quality was also determined and found that, pH of breast meat left from slaughter 45 minutes (pH45) and 24 hours (pH24) of broiler was significantly higher than Thai Native and Betong (KU line) ($P < 0.01$) whereas the percentage of drip loss, thawing loss, cooking loss and shear force was not significantly different between breeds. The polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) technique was used to genotype the polymorphism in the *ApoVLDL-II* gene in the broiler, Thai Native and Betong (KU line) chickens. The results found that, the polymorphism in the *ApoVLDL-II* gene at VLDL6 loci was not associated with fat accumulation in those studied population.

Keywords*—ApoVLDL-II* Gene, Betong (KU line) chickens, broiler chickens, carcass characteristic, growth performance, meat quality, Thai Native Chickens.

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I. Introduction

THE Betong chicken (KU Line), a slow growing chicken is a meat type strain popular in the southern region of Thailand. The Betong produces a high-quality meat which is softer and has a better taste that of other native Thai (low carcass fat and high lean meat(, but is not as flabby as broiler's meat [1]. Thai native chickens are raised by small farmers and is a slow growing chicken with poor performance. However, both Betong chicken (KU Line) and Thai Native chickens were the high quality of the meat and low carcass fat compared to broiler chickens. In Thailand, with the increasing demand for chicken meat, farm households tend to produce more broilers and other Thai Native chicken breeds, especially on a large scale. However, compared to commercial broilers, Betong chicken (KU Line) and Thai Native chicken have not been produced enough for consumer demand because of their poor growth performances and little is known about the meat quality of Thai Native breeds. To understand fat accumulation in chickens, the objectives of the present study were to identify the polymorphisms of the chicken *ApoVLDL-II* (Apo-very low density lipoprotein II) gene in the broiler, Thai Native and Betong (KU line) chickens. In growing chickens, very low-density lipoprotein (VLDL) is is the major transporter of triglycerides and attempts to reduce excessive fatness in poultry. Moreover, the VLDL genes reported most abundant mRNA species present in livers of hens or estrogen treated roosters [2]. We developed PCR-RFLP to genotype the polymorphisms of *apoVLDL-II* gene and study the growth performance, carcass characteristics and meat quality of the broiler, Thai Native and Betong (KU line) chickens.

II. Materials and Methods

A. Experimental Stocks and Growth Performance

The female broiler, Thai Native and Betong (KU line) chickens' chicken were reared under the same environment and management from 4-14 weeks (100 chicks per breed). In order to determine weight gain, all birds were weighed in the beginning, every 2 weeks, and the end of the experimental period. These data were used to measure BW, average daily gain (ADG) and feed intake (FI). FCR was calculated from

BW and FI. At 14 weeks of age, all chickens were euthanized by CO_2 inhalation. Immediately after euthanasia, these birds were killed and slaughtered. Carcass and organs were reported as a percentage of live weight.

B. Meat Quality

To determine breast meat quality, the breast muscle (*M. pectoralis*) was dissected from each broiler chicken sample and evaluated for meat quality characteristics. Meat quality measurements include drip loss, thawing loss and cooking loss, and are calculated as the difference between initial and final weight, and expressed in percentage. Shear force data were obtained from TA-XT. plus texturometer and analyzed by a specific software (Stable Microsystems Ltd., Surrey, UK). The pH was measured in the left breast muscles 45 minutes (pH_{45}) after slaughter and after 24 hours (pH_{24}) of cooling at 4 ºC, using spear tip electrode combined with pH-meter (CP-401, ELMETRON, Poland).

C. DNA Isolation and PCR-RFLP

At 14 weeks of age, 0.5 ml blood samples were taken from the wing vein of respective populations for DNA extraction. Genomic DNA was isolated from the whole blood using saturated salt method [3]. PCR-RFLP amplification of *ApoVLDL-II* primer (Forward 5'- CCT CTA TGA CAT GGT TGC CT-3'; Reverse 5'- ATG GGT TTG ACC CTG CTA TG-3') was carried out in total volume of 25 µl, It was initiated with a first denaturation step of 5 min at 95 °C, followed by 40 cycles of 94 °C for 2 min and 55 °C for 1 min. PCR products were checked in 1.0% agarose gels stained with ethidium bromide to determine the presence of product. Thereby, 10 µl of each PCR products were digested overnight at 37 °C by 3 U of *SfcI* restriction enzyme (Thermo Scientific, USA). The digested fragments were electrophoresed using 2% agarose gels stained with ethidium bromide to determine the presence of product.

D. Statistical Analysis

The least square model according to the characteristic of experimental material was constructed. Analysis software is SAS (version 9; SAS Institute Inc., Cary, NC, USA)

III. Results and Discussion

The growth performance of broiler, Thai Native and Betong (KU line) chickens were shown in Table I and II. The BW of broilers was highest)P < 0.01(in all experimental periods. The BW of Thai Native was higher than Betong (KU line) chickens (P < 0.01).

TABLE I
Least Square Means and Standard Error of BW and Average Diary Gain of the Female Broiler, Thai Native and Betong (KU Line) Chickens

Items	Groups			P-value
	Broilers	Thai Natives	Betong	
BW♯				
4 wks	1,255.70 ± 3.41[a]	366.47 ± 3.41[b]	321.31 ± 3.41[c]	<.001
6 wks	2,246.22 ± 9.35[a]	602.99 ± 9.21[b]	539.92 ± 9.21[c]	<.001
8 wks	3,113.02 ± 66.65[a]	872.28 ± 63.53[b]	781.82 ± 62.55[b]	<.001
10 wks	3,450.22 ± 147.65[a]	999.95 ± 132.35[b]	969.13 ± 115.85[b]	<.001
12 wks	3,739.14 ± 178.36[a]	1,244.63 ± 92.55[b]	1,141.19 ± 82.01[b]	<.001
14 wks	3,974.22 ± 118.02[a]	1,459.84 ± 59.93[b]	1,332.80 ± 50.77[b]	<.001
ADG♯				
4 wks	49.70 ± 0.17[a]	15.11 ± 0.17[b]	11.99 ± 0.17[c]	<.001
6 wks	70.80 ± 0.64[a]	16.98 ± 0.63[b]	15.65 ± 0.63[b]	<.001
8 wks	63.36 ± 3.96[a]	19.00 ± 4.23[b]	17.21 ± 3.76[b]	<.001
10 wks	26.04 ± 6.17	13.59 ± 6.36	13.56 ± 4.88	0.282
12 wks	18.46 ± 4.54	18.50 ± 1.99	12.27 ± 1.59	0.084
14 wks	20.20 ± 4.33	15.99 ± 2.17	13.72 ± 1.73	0.377

ADG (g/h/d). [a, b, c] Means in the same row with different superscript are significantly difference (P < 0.01).

In broiler chickens, the FI was higher than Thai Native was higher than Betong (KU line) chickens (P < 0.01) at 4-10 weeks, while there was no significant difference between breeds at 10-14 weeks. Based on our results, the broilers showed better FCR compared to other breeds)P < 0.01(at 4-8 weeks.

The carcass characteristics of the female Broiler, Thai Native and Betong (KU line) chickens are shown in Table III.

There were no significant differences between breeds in the total percentage carcass yield and drumstick)P > 0.01(. The highest values of breast muscle and tenderloin percentages were found in the group of broiler chickens (P < 0.01), whereas there was no significant difference between groups of Thai Native and Betong (KU line) chickens.

The percentage of wing was higher in Thai Native and Betong (KU line) chickens than broilers. Relative weights of liver were highest in Thai Native chicken, whereas the value was similar between the broilers and Betong chicken.

The percentage of abdominal and subcutaneous fat in Thai Native and Betong (KU line) chickens were significantly lower than the broilers (P < 0.01). These results are in agreement with the findings of [4] and [5], who reported a similar growth performance of a slow growing Betong chicken (KU Line). Moreover, the previous studies reported a slowing growth and carcass of Thai Native chickens [6], [7].

TABLE II
LEAST SQUARE MEANS OF FI AND FCR OF THE FEMALE BROILER, THAI NATIVE AND BETONG (KU LINE) CHICKENS

Items	Groups			SEM	P-value
	Broilers	Thai Natives	Betong		
FI					
4-6 wks	134.41[a]	38.48[b]	37.76[b]	4.19	<.001
6-8 wks	124.89[a]	49.88[b]	47.86[b]	7.67	<.001
8-10 wks	95.74[a]	46.55[b]	45.41[b]	8.29	0.002
10-12 wks	94.89[A]	60.40[B]	45.39[B]	9.59	0.015
12-14 wks	79.37	56.64	54.96	11.73	0.307
FCR					
4-6 wks	1.89[b]	2.26[a]	2.41[a]	0.07	0.002
6-8 wks	1.97[B]	2.60[A]	2.78[A]	0.19	0.035
8-10 wks	5.18	3.52	3.35	0.91	0.339
10-12 wks	3.92	3.29	3.70	0.43	0.601
12-14 wks	3.33	3.37	4.00	0.53	0.619

FI (g/h/d); [a, b, c] Means in the same row with different superscript are significantly difference (P < 0.01).

TABLE III
LEAST SQUARE MENAS AND STANDARD ERROR OF CARCASS CHARACTERISTICS OF FEMALE BROILER, THAI NATIVE AND BETONG (KU LINE) CHICKENS (PERCENTAGE OF LIVE WEIGHT)

Items	Groups			P-value
	Broilers	Thai Natives	Betong	
Carcass	84.17 ± 0.95	82.50 ± 0.52	82.25 ± 0.42	0.243
Breast muscle	15.37 ± 0.50[a]	8.20 ± 0.27[b]	7.81 ± 0.22[b]	<.001
Drumstick	19.81 ± 0.45	21.15 ± 0.24	20.94 ± 0.20	0.086
Tenderloin	4.42 ± 0.13[a]	3.75 ± 0.07[b]	3.47 ± 0.05[b]	0.006
Wing	7.61 ± 0.16[b]	9.44 ± 0.89[a]	9.70 ± 0.07[a]	<.001
Liver	1.21 ± 0.06[b]	1.56 ± 0.03[a]	1.35 ± 0.02[b]	0.001
Abdominal fat	4.78 ± 0.49[a]	1.29 ± 0.24[b]	2.07 ± 0.20[b]	0.003
Subcutaneous fat	3.68 ± 0.53[a]	0.79 ± 0.30[b]	1.39 ± 0.22[b]	0.004

[a, b, c] Means in the same row with different superscript are significantly difference (P < 0.01).

Regarding fat content, the highest fat content was recorded in broilers. Similar results have recently been observed, where broilers had a higher fat content in both meat portions than Thai Native breeds [8]. Thai native chickens also included a lower fat than broilers and unique compositional characteristic. In this study, however, broilers had more fat in the meat than the older slow-growing birds. This confirms that the genotypic effect on fat deposition could go beyond age influence in some cases [9]. Additionally, we found the increasing of growth rate in broiler chickens has been associated with increased fat deposition [10].

In terms of the pH of breast meat, Thai Native and Betong chicken showed lower values than all other types of chicken investigated in the present study at pH_{45} and pH_{24} (P < 0.01; Table IV). The highest pH value was observed in the breast meat of broilers. Differences in pH values among the breeds of chicken investigated may be attributed to the pre-slaughter stress, which changes muscle glycogen content and eventually has an effect on the rate and extent of pH decline [11]

In our study, the percentage of drip loss, thawing loss, cooking loss and shear force were similar among the breeds of chicken (Table IV).

TABLE IV
LEAST SQUARE MEANS AND STANDARD ERROR OF MEAT QUALITY OF THE FEMALE BROILER, THAI NATIVE AND BETONG (KU LINE) CHICKENS

Items	Groups			P-value
	Broilers	Thai Natives	Betong	
pH_0	6.57 ± 0.17	6.10 ± 0.07	6.15 ± 0.06	0.124
pH_{45}	6.40 ± 0.10[a]	5.84 ± 0.06[b]	5.91 ± 0.04[b]	0.004
pH_{24}	5.83 ± 0.03[a]	5.66 ± 0.01[b]	5.70 ± 0.01[b]	0.006
Drip loss[1]	5.53 ± 1.15	4.86 ± 1.05	5.13 ± 0.53	0.913
Thawing loss[1]	4.46 ± 1.01	3.17 ± 0.98	4.68 ± 1.13	0.579
Cooking loss[1]	16.91 ± 1.55	14.24 ± 1.50	13.03 ± 1.72	0.323
Shear force[2]	2.49 ± 0.14	2.36 ± 0.13	2.70 ± 0.10	0.861

[1] Percentage of Drip loss, Thawing loss and Cooking loss; [2] Shear force)kg/cm^2(; [a, b, c] Means in the same row with different superscript are significantly difference (P < 0.01).

In this study polymorphism of *ApoVLDL-II* gene at VLDL6 loci were performed by PCR-RFLP (Fig. 1), the amplified 492-bp product from the *ApoVLDL-II* gene. For total 300 chickens of the female broiler, Thai Native and Betong (KU line) chickens, we found only genotype BB (396 bp and 96 bp). The results found that, the polymorphism in the *ApoVLDL-II* gene at VLDL6 loci was not associated with fat accumulation in those studied population.

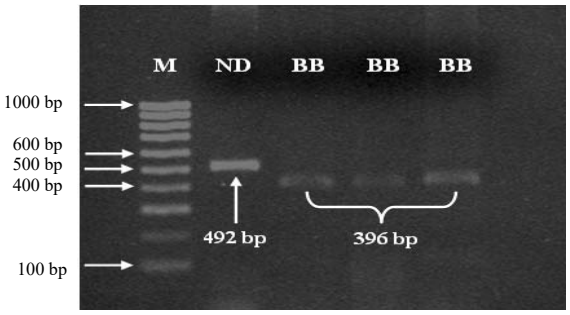

Fig. 1 The PCR-RFLP pattern for the *ApoVLDL-II* gene with *SfcI* digestion. M = Marker; ND (no digestion PCR product) = 492 bp; BB = 392, 96 bp. The numbers listed on the left of the figure are fragment sizes

However, the study of lean and fat chicken lines found heterozygous genotype in VLDL6 locus (AA) observed significantly (P<0.05) higher BW and fat weight [12], [13] also reported other locus of *ApoVLDL-II* gene in lean and fat chicken lines including VLDL9, VLDL10 and VLDL17. They found polymorphism in apoVLDL-II gene was significantly (P < 0.05) associated with BW and fat weight at VLDL9 and VLDL17 loci in lean chicken. In addition, polymorphism of *apoVLDL-II* gene at VLDL6, VLDL9 and VLDL10 loci was significantly (P < 0.05) associated with BW and fat weight. Moreover, [13] reported the interaction of *ApoVLDL-II* gene and lipoprotein lipase (LPL) gene with fat in lean and fat chicken breeds. These finding suggested that growth and fat accumulation were control by many loci of *apoVLDL-II* gene. Furthermore, in chickens reported that the polymorphysim and expression of lipogenic enzyme regulated fat deposition during chicken adipogenesis [14], [15] reported that fatty acid synthase, suggest the possibility that a combination of malic

enzyme, fatty acid synthase or acetyl CoA carboxylase and lipoprotein lipase, which gene polymorphism and mRNA expression interact to regulate lipogenesis in the chicken.

IV. Conclusion

In conclusion, we found that growth rate of Thai Native and Betong chickens (KU Line) are slower than broilers, while the lipogenic capacity of adipose tissue is decreased in Thai Native and Betong chickens (KU Line). Meat quality was not significantly different between breeds. The polymorphism in the *ApoVLDL-II* gene at VLDL6 loci was not associated with fat accumulation in those studied population.

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