Manipulation of Very Low Density Lipoprotein Metabolism to Reduce Fat Deposition in Poultry

Loh, TC.[1], Tan, BK.[1], Foo, HL.[2], Abdullah, N.[3], Idrus, Z.[1] and Ramlah, H.[1]

[1]Department of Animal Science,
Faculty of Agriculture
[2]Department of Biotechnology, [3]
Department of Biochemistry,
Faculty of Science and Environmental Studies
University Putra Malaysia,
43400 UPM Serdang, Selangor.
Malaysia

Telephone Number of Corresponding Author: 03-8946 6899

E-mail of Corresponding Author: tcloh@agri.upm.edu.my

Introduction

In birds, lipids (especially triacylglycerol) are mainly deposited in adipocyte, hepatocytes and growing oocytes (i.e. vitellogenesis). Excessive accumulation of lipids in the adipose tissues of modern lines of broilers is a major concern because fat is expensive to produce (due to depress feed efficiency), thrown out as an offal (due to eviceration) and less appreciated by consumer (due to poor dietetic quality). Since fat has many drawbacks, its excessive accumulation must be limited. *De novo* lipogenesis is very limited in bird's adipose tissue. Storage of triacylglycerol in this tissue depends on the availability of plasma lipid originating either from the diet or lipogenesis in the liver. A better understanding of the metabolism and biochemistry of lipoprotein in association to fat deposition would be of considerable value in carcass quality. It could also have a profound effect on the productivity of poultry farming. The objectives of this project were to study the relationships among plasma, VLDL, VLDL subfractions lipids, postheparin plasma lipoprotein lipase (LPL) activity and fat deposition in two different breeds of chickens and to characterise their VLDL apolipoprotein.

Materials and Methods

A total of 720 1-day-old commercial broiler (Avian), (CB) and crossbred village chicken (village chicken x Sasso), (AK) were used in this experiment. Chicks were reared in floor pens (3.72m^2) with wood shaving as litter and provided conventional starter (day 1 to day 14) and finisher diet (day 15 onwards) ad libitum. The chicks were housed in groups of 60 until 6 weeks for CB and 12 weeks for AK. Weekly feed intake and body weight were recorded. Blood and abdominal adipose tissue were collected at 3 weeks interval. The chicks were fasted overnight before collection of samples. Blood was collected into a vacutainer tube containing EDTA as anticoagulant. Plasma was isolated by centrifuging at 1500g for 30 minutes. Plasma VLDL was then purified by using Fast Protein Liquid Chromatography (FPLC) at a flow rate of 0.4ml/min. 0.15M NaCl, 10mM Na_2HPO_4, 5mM Na_2EDTA and 0.02% NaN_3 were used as running buffers. Lipid compositions in plasma and VLDL were determined by using the respective diagnostic kits (TAG and cholesterol from Randox, free cholesterol and phospholipid from Wako). Abdominal adipose tissues were collected in an ice box and weighed. The trial was set up in a randomized block design. Data were analyzed by analysis of variance (ANOVA) using the general linear model (SAS, 1988). A least significant difference (LSD) was used to compare means differences.

Results and Discussion

In gel filtration chromatography, the molecules are eluted with decreasing particle size. Peak A may represents VLDL as the particle size is the largest among the lipoproteins. In order to prove that, peak A fractions were pooled and analysed with TEM. The average particle size is 46.8nm. For further confirmation, PAGE is performed (Figure 3) with 4% gel. The VLDL particle weight is believed to be between 40,000,000 and 5,000,000 daltons. However, there is no such big molecular weight marker in the market. Therefore, the whole chicken plasma was used to compare the electrophoresis gel pattern. Lanes 1 and 4 are the crossbred village chicken (VC) and commercial broiler (CB) plasma samples, respectively. Lane 2 represents peak A from the VC plasma sample whereas lane 3 is from the CB plasma sample. Two bands were obtained in VC (lane 1) and CB (lane 4) plasma samples. First and second bands are the VLDL and LDL, respectively. High density lipoprotein had been eluted from the gel as the molecular size is too small to be retained in 4% PAGE gel. Only one band is obtained in VC (lane 2) and CB (lane 3) FPLC fractions and these bands are parallel with the first band in plasma sample. These results indicate that peak A is VLDL. Commercial broiler deposited significantly ($P<0.001$) more fat than AK at weeks 3 and 6. This is agreed with Whitehead et al. (1990) study, who claimed that fat line broiler deposited higher fat than lean line broiler. Plasma TAG concentration for CB was significantly lower ($P<0.05$) than AK chicken at week 3. However, VLDL TAG concentration was not statistically significant ($P>0.05$) between these two lines. Plasma ($P<0.001$) and VLDL ($P<0.05$) TAG for CB were significantly lower than AK at week 6. This could be explained by either a decrease in secretion of TAG from the liver into the circulation or an increase in uptake of TAG from the blood into the adipose tissue and thus resulted in a lower concentration of TAG in plasma. Additionally, there was a significant negative relationship between VLDL TAG concentration and fat deposition at week 3 ($R=-0.45$, $P<0.05$) but not at week 6 ($R=-0.22$, $P>0.05$). These results are in disagreement with the findings of Griffin et al. (1982), who reported that birds with higher plasma TAG concentration were significantly fatter than

those with lower plasma TAG concentration. This could be explained by the different methods of collecting and analyzing samples. In the present study, the birds were fasted overnight before collecting samples and VLDL was purified by using FPLC according to size exclusion principle. In the experiment conducted by Griffin et al. (1982), the birds were not fasted before blood collection and the VLDL was isolated from plasma by precipitation method using dextran sulphate and $MnCl_2$. Excessive portomicrons will be present in the blood circulation if the birds are not fasted. This may give rise to errors in the determination of TAG level in VLDL as portomicrons and VLDL have similar density.

Conclusions

The body weight (BW) and feed intake (FI) of CB were significantly higher than that of AK but the feed conversion ratio was significantly lower. CB showed a significant higher proportion of subfraction 2, bigger VLDL particle size and higher postheparin plasma LPL activity than AK. All these lead to a higher fat deposition of CB than that of AK.

Benefits from the study

A clear description of the changes to lipoprotein compositions occurring during growing period was obtained. These changes are important to the metabolism of the lipoproteins and their implications for manipulation of carcass fat deposition rate in meat animals

Patent(s), if applicable :

Nil

Stage of Commercialization, if applicable :

Nil

Project Publications in Refereed Journals

1. Loh, T.C., H.L. Foo, Ramli, M. and Tan, B.K. (2002) Effect of dietary chromium picolinate on abdominal fat and lipid metabolism of broilers. *Online Journal of Veterinary Research.* 1: 53-58.

2. Tan, B.K.,'H.L. Foo, T.C. Loh, Abdullah, N., Idrus, Z. Characterisation and purification of very low density lipoprotein by fast fast protein liquid chromatography. Journal Chromatography A (submitted)

Project Publications in Conference Proceedings

1. Loh, T.C., B.K.Tan, H.L. Foo, Idrus, Z., Abdullah, N. and Ramlah, H. (2002) Blood lipids and fat depsoiton in commercial broiler and crossbred village chicken. Inventions and Research 2002, Faculty of Agriculture. pp. 123-126.

2. Tan, B.K., T.C. Loh, H.L.Foo, Zulkifli, I. and N. Abdullah (2001) Bloods lipids and fat deposition in commercial broiler and crossbred village chicken. UPM Research Report 2001: Faculty of Agriculture. pp. 42-45.

3. Tan, B.K., T.C. Loh, H.L. Foo, Idrus, Z. and Abdullah, N. (2000) Blood lipids and fat deposition in commercial broiler and crossbred village chicken. In*: Animal food production, future vision.* pp: 180-182.

4. Tan, B.K., T. C. Loh, H. L. Foo, Abdullah , N., Idrus, Z. and P.F. Dodds (2000) Purification of very low density lipoprotein from chicken's plasma by fast protein liquid chromatography. In *Biotechnology as the catalyst for economic growth: from genes to proteins.* 12th National Biotechnology Seminar. pp: 20-22.

Graduate Research

Name of Graduate	Research Topic	Field of Expertise	Degree Awarded	Graduation Year
Tan Bee Koon	Very low density lipoprotein metabolism and fat depostion of commecial broiler and crossbred village chicken	**Animal Biochemistry**	**MS**	**2002**

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