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学位の種類	博士（医学）
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審査研究科	人間総合科学研究科
学位論文題目	RNA-sequencing analysis of paternal low-protein diet-induced gene expression change in mouse offspring adipocytes（父親への低タンパク質餌による次世代マウス脂肪細胞での遺伝子発現変化の RNA シーケンシング解析）
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論文の内容の要旨
Abstract of thesis

In this doctoral dissertation, Ms. Ly describes the effect of paternal low-protein diet for gene expression in inguinal white adipose tissue of offspring. The summary is as follows:

（目的　Purpose）

Increasing evidence indicates that parental diet affects the metabolism and health of offspring. It is reported that paternal low-protein diet induces glucose intolerance and the expression of genes involved in cholesterol biosynthesis in mouse offspring liver. The aim of the present study was to determine the effect of a paternal low-protein diet on gene expression in offspring inguinal white adipose tissue, another important tissue for the regulation of metabolism. The author also compared gene expression pattern between the liver and inguinal adipose tissue, and characterize the difference between generations and tissues.

（対象と方法　Materials and Methods）

To determine the effects of a paternal low-protein diet on gene expression in offspring inguinal white adipose tissue, the author used essentially the same feeding regimen in the previous reports. In brief, after weaning at 3 weeks of age, male F0 mice were fed the control diet or low-protein diet, then mated with female F0 mice which were fed the control diet to produce F1 offspring. Transcriptomic profile of inguinal white adipose tissue from male F1 offspring at 5 weeks of age were analyzed by RNA-seq. To see whether the alteration in gene expression pattern in F1 offspring inguinal white adipose tissue mimicked that in their male parents, the author performed transcriptome

profiling of F0 inguinal white adipose tissue in 10-12 weeks old male mice fed the control diet or low-protein diet and compared transcriptome profiles in inguinal white adipose tissue between two generations. Finally, to understand how paternal low-protein diet induced differences in gene expression in the inguinal white adipose tissue and liver of the offspring contribute to the glucose intolerance that was previously reported, the author also compared the transcriptome between 2 tissues, using the differentially expressed genes in F1 liver, which was previously analyzed following a similar feeding study.

（結果　Results）

Paternal low-protein diet up- and down-regulated 54 and 274 genes, respectively, in offspring inguinal white adipose tissue. The down-regulated genes contained numerous representatives from metabolic pathways, such as fatty acid biosynthesis and glycolysis. The expression of carbohydrate response element-binding protein β (ChREBP-β), an important lipogenic transcription factor, was markedly reduced in paternal low-protein diet offspring. Paternal low-protein diet reduced the amount of ChREBP, presumably ChREBP-α, binding to the promoter of ChREBP-β, leading to a reduction in the expression of ChREBP-β. Moreover, Igfbp2 was among most up-regulated genes, which may inhibit glycolysis, leading to suppression of the nuclear translocation of ChREBP-α. Furthermore, the analysis of the differentially expressed genes of F0 inguinal white adipose tissue showed that lipolysis is up-regulated in F0 inguinal white adipose tissue. Only 12 genes showed overlapping differential expression among the 274 and 334 down-regulated genes in the F1 and F0 inguinal white adipose tissue, respectively. Comparison of the differentially expressed genes in F1 inguinal white adipose tissue and liver showed almost no overlap, with only one and five genes overlapping among the up- and down-regulated lists, respectively.

（考察　Discussion）

Carone *et al*. demonstrated that paternal low-protein diet increased the expression of many cholesterol biosynthesis-regulating genes in offspring liver. Using the same diet and feeding regimen, the author found that paternal low-protein diet down-regulates multiple lipogenic genes in offspring inguinal white adipose tissue. Both of these opposite effects on lipogenesis of liver and inguinal white adipose tissue would be expected to cause insulin resistance. It is as yet unclear how a paternal low-protein diet might differentially affect the expression of genes in these two offspring tissues. Low-protein diet may induce an epigenetic change, such as histone modification or RNA induction, in the testicular germ cells, which may be transmitted to the mature sperm. Because differentiation into tissues such as liver and inguinal white adipose tissue occurs in the post-blastocyst embryo, such changes in the early embryo would be expected to affect gene expression in the embryo at a later stage. A low-protein diet up-regulates the expression of lipid oxidation genes in the inguinal white adipose tissue of F0 mice. The author discusses that differences in gene expression in offspring inguinal white adipose tissue may be the result of epigenetic changes in the testicular germ cells of their male progenitors. The author considers that further analysis is required to dissect the mechanism underlying the differential effects of a paternal low-protein diet on the expression of genes in inguinal white adipose tissue and liver.

審査の結果の要旨
Abstract of assessment result

（批評　General Comments）

The author described that mRNA expression of many genes involved in lipogenesis was down-regulated by paternal low-protein diet feeding in inguinal white adipose tissue, which may contribute to glucose intolerance. The expression of ChREBP-β was also significantly lower in the inguinal white adipose tissue of paternal low-protein diet offspring, which may have mediated the down-regulation of the lipogenic genes. This analysis adds new information about epigenetic effect of nutrition.

（最終試験の結果　Assessment）

The final examination committee conducted a meeting as a final examination on May 30, 2019. The applicant provided an overview of dissertation, addressed questions and comments raised during Q&A session. All of the committee members reached a final decision that the applicant has passed the final examination.

（結論　Conclusion）

The final examination committee approved that the applicant is qualified to be awarded Doctor of Philosophy in Medical Sciences.