京都大学	博士 (ゲノム医学)	氏 名	Satoshi Yoshiji
論文題目	Leveraging genomics and proteomics to identify therapeutic targets for COVID-19 and cardiometabolic diseases (ゲノム・プロテオーム解析を用いた COVID-19 および心血管代謝疾患の創薬標的同定)		

（論文内容の要旨）

Obesity is a major risk factor for coronavirus disease 2019 (COVID-19) severity and cardiometabolic diseases, including coronary artery disease (CAD), stroke, and type 2 diabetes. However, the underlying mechanisms through which obesity influences these diseases are not fully understood. By leveraging genomics and proteomics, this thesis investigates how circulating proteins mediate the effect of obesity on COVID-19 severity and cardiometabolic diseases.

First, the causal effect of obesity on COVID-19 severity were evaluated using Mendelian randomization (MR), a causal inference method in genetic epidemiology. Given that body fat mass and fat-free mass are genetically interrelated, multivariable MR were utilized to discern their independent causal effects. The findings showed that body fat mass is independently associated with an increased risk of COVID-19 severity.

Second, considering that obesity strongly influences the plasma proteome, this thesis sought to identify circulating proteins that mediate the effect of obesity on COVID-19 severity. Proteome-wide MR were used to estimate the causal effect of BMI on circulating protein levels and identified proteins whose plasma levels are influenced by BMI, termed "BMI-driven proteins". Then, the causal effects of the BMI-driven proteins on COVID-19 outcomes were evaluated, again using MR with cis-acting protein quantitative trait loci (cis-pQTLs). This two-step MR approach found that increased circulating nephronectin (NPNT) levels were associated with an increased risk of critically ill and COVID-19 hospitalization. To ensure the robustness of the findings, the analyses were repeated using body fat percentage and cis-pQTLs from different cohorts, which consistently showed that NPNT partially mediates the effect of obesity on COVID-19 severity. In further follow-up analyses for NPNT, a specific NPNT isoform was found to drive the effect. In single-cell RNA sequencing of the lung from individuals who died of COVID-19, NPNT was significantly expressed in fibroblasts and alveolar cells. Finally, multivariable MR revealed that decreasing body fat mass and increasing fat-free mass can lower NPNT levels and thus may improve COVID-19 severity—underscoring NPNT's potential clinical relevance and actionability.

Finally, this thesis expanded this framework to other major complications of obesity: CAD, stroke, and type 2 diabetes. Using the two-step MR approach, followed by colocalization and mediation analysis, seven plasma protein mediators with eight protein-disease associations were identified. Among them, circulating collagen type VI alpha-3 (COL6A3) was strongly increased by BMI and increased the risk of CAD.

In follow-up analysis for COL6A3, the causal effect of its C- and N-terminal effects on CAD was evaluated. This domain-aware MR found that the C-terminal fragment of COL6A3, known as endotrophin, mediated the effect. In single-cell RNA sequencing of adipose tissues and coronary arteries, COL6A3 was highly expressed in cell types involved in metabolic dysfunction and fibrosis. Finally, multivariable MR revealed that body fat reduction can lower plasma levels of COL6A3-derived endotrophin and other protein mediators and reduce the risk of cardiometabolic diseases.

In summary, this thesis provides clinically relevant insights into how circulating proteins mediate the effect of obesity on COVID-19 severity and cardiometabolic diseases. This integrative proteogenomics approach prioritizes potential therapeutic targets, including NPNT for COVID-19 and endotrophin for CAD.

（Summary of Dissertation Examination Results）

Obesity is a strong risk factor for severe COVID-19 and cardiometabolic disorders, such as coronary artery disease (CAD), stroke, and type 2 diabetes. Yet, the underlying mechanisms remain unclear. This thesis explored how circulating proteins influence the effect of obesity on COVID-19 severity and cardiometabolic diseases using genomic and proteomic approaches.

First, the thesis assessed the direct causal relationship between obesity and COVID-19 severity through Mendelian randomization (MR), disentangling their causal impacts. The results indicated that body fat mass, but not fat-free mass, independently increases the risk of severe COVID-19.

Second, the thesis used proteome-wide MR to reveal that plasma nephronectin (NPNT) mediates obesity's effect on COVID-19 risk, with a particular NPNT isoform being the key driver. NPNT is significantly expressed in fibroblasts and alveolar cells in COVID-19-affected lungs. Additionally, multivariable MR suggests that decreasing body fat mass could reduce NPNT levels and thus reduce the risk of COVID-19 severity.

Lastly, the thesis also explores how obesity increases the risk of cardiometabolic diseases. The two-step MR approach pinpointed seven plasma

proteins as mediators, including collagen type VI alpha-3 (COL6A3). Interestingly, the C-terminal fragment of COL6A3, known as endotrophin, was found to mediate this effect. Single-cell RNA sequencing of adipose tissues and coronary arteries showed that COL6A3 was highly expressed in cell types involved in metabolic dysfunction and fibrosis. Further, multivariable MR revealed that body fat reduction could lower plasma levels of endotrophin and reduce the risk of cardiometabolic diseases.

Overall, the thesis offers valuable clinical insights into the role of circulating proteins in linking obesity to COVID-19 severity and cardiometabolic diseases. Its integrative proteogenomics approach highlights potential therapeutic targets, including NPNT for COVID-19 and endotrophin for CAD.

From the very convincing answers of the candidate to the numerous questions of the jury, it is clear that he has a deep understanding of a wide range of topics related to genetic epidemiology, genomics, proteomics, COVID-19, and cardiometabolic diseases.
The answers also revealed that he has an active and already well advanced ongoing research project, beyond the results of the thesis. As a result of the oral examination held on 2023/12/11 regarding the dissertation contents and related matters, we certify that the candidate has passed the oral examination.