1. No.
   2023G001

2. Title
   Genetic Metabolic Diversity of CYP2A6 a Nicotine-Metabolizing Enzyme Using a General Population Biobank

3. Period
   2023-2025

4. Summary

This study aimed to elucidate the genetic metabolic diversity of cytochrome P450 2A6 (CYP2A6), a key nicotine-metabolizing enzyme involved in the metabolic activation of tobacco-derived carcinogenic precursors, using large-scale population biobank data. First, we established an improved mammalian cell expression system that enabled higher expression and activity of CYP2A6 by optimizing cofactor supplementation (iron ions and 5-aminolevulinic acid) and co-expression of electron transfer partners, including NADPH-cytochrome P450 reductase and cytochrome b5. In addition, we developed a high-throughput functional evaluation system using the fluorescent probe substrate coumarin, which significantly accelerated the analysis of CYP2A6 variants. Using these systems, we comprehensively evaluated the functional effects of rare CYP2A6 variants identified from Japanese whole-genome sequencing data. Enzyme kinetic analyses revealed several variants with markedly reduced or abolished catalytic activity compared with the wild-type enzyme. These findings provide important insights into the genetic determinants underlying interindividual variability in nicotine metabolism and susceptibility to smoking-related carcinogenesis and establish a functional framework for evaluating rare pharmacogenetic variants in population genomic data.