Plasma ^{1}H NMR-based metabolomic analysis of the lipid regulating effect of novel carboxamide derivatives in Triton WR-1339 induced hyperlipidemic rats

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Abstract

Hyperlipidemia is a heterogenous metabolic disorder defined as an abnormal increase in circulating levels of one or more plasma lipids and lipoproteins including total cholesterol, triglycerides, and low-density lipoprotein-cholesterol and/or reduction of high-density lipoprotein-cholesterol. Hyperlipidemia is the main risk factor of cardiovascular diseases which are considered the primary cause of morbidity and mortality worldwide. This study aimed to identify potential biomarkers for hyperlipidemia. It also aimed to explore the underlying mechanism of the metabolic disturbances, and to investigate the lipid lowering effect of carboxamide compounds (X, Y, and Z) via integrating NMR-based metabolomics and transcriptomics. Hyperlipidemia was induced in rats by the

administration of Triton WR-1339. Carboxamide compounds (X, Y, and Z) were administered intragastrically and their activity was compared to the lipid lowering agent fenofibrate, due to their similar functional groups. Plasma samples were collected for lipid profile assay and metabolic profiling. Plasma samples were metabolically profiled with nuclear magnetic resonance spectroscopy (NMR) using cryogenic probe. Liver, cardiac, kidney and endothelial tissues were collected for gene expression analysis by RT-PCR. The three tested carboxamide compounds were able to significantly reduce the elevated plasma triglycerides levels. Metabolomics results revealed 12 metabolites as potential biomarkers associated with hyperlipidemia which are NAD+, succinate, inosine, cysteine, phenylalanine, tyrosine, serine, glycine, myo-inositol, glucose, ATP and creatine phosphate. Elevated levels of the potential biomarkers were observed in the plasma of hyperlipidemic rats and these potential biomarkers are mainly associated with inflammation, oxidative stress and abnormal lipid metabolism. Administration of carboxamide compounds restored the levels of metabolites and reversed the metabolic disturbance in plasma of hyperlipidemic rats. Metabolomic results were correlated with the expression of related genes in different tissues. Gene expression results revealed the underlying mechanism of metabolites levels elevation in hyperlipidemia by significantly altering the expression of *SLC29A1*, *SLC2A2*, *SLC16A10*, *SDHA*, *SLC25A51* in the liver, altering *SLC29A1*, *SLC25A10* and *SLC5A3* in cardiac tissue, and altering *SLC16A1* in kidney tissue. Carboxamide compounds were able to restore gene expression levels of many related genes, thus validating the metabolomics results and revealing a proposed mechanism in which carboxamide compounds ameliorate hyperlipidemia induced metabolic disturbance. Our findings demonstrate the mechanism underlying the

development of hyperlipidemia and the potential activity of carboxamide compounds as lipid lowering compounds.

Keywords: Carboxamide derivatives, Hyperlipidemia, Metabolomics, NMR spectroscopy, Potential biomarkers.