Notch ligand Delta-like 4 blockade attenuates atherosclerosis and metabolic disorders

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AUTHOR SUMMARY

Obese people have a greater risk of death from all causes, which is accounted for by atherosclerosis-induced cardiovascular disease. A disease complex called “cardiometabolic syndrome” is characterized by atherosclerosis, obesity, insulin resistance, and fatty liver. This disease has been linked to chronic inflammation, in which a key role is played by macrophages that are more abundant in atherosclerotic lesions than in healthy arteries. Fat tissue of obese individuals contains more macrophages than does the fat tissue of healthy-weight individuals. We know that in cultured macrophages a protein called Notch ligand delta-like 4 (Dll4) triggers various proinflammatory effects associated with atherosclerosis and metabolic disorders (1), and our goal in the present study was to replicate these findings in animals. Here, we report that suppression of Dll4 simultaneously attenuates several key components of cardiometabolic syndrome in a mouse model system.

Exercise and healthy diets are generally the first choice for the prevention or treatment of metabolic diseases. Many at-risk individuals’ low adherence to therapeutic lifestyle changes, however, has driven the necessity of effective medical therapies to combat cardiometabolic syndrome, the global epidemic. Strong evidence for the role of inflammation in atherosclerosis and metabolic disorders led to our belief that if investigators who study inflammation share their knowledge across different disease contexts, we will understand better how inflammation associates with disorders in multiple metabolic organs in parallel. This collaboration promises to provide unique insight into the development of well-defined antiinflammatory therapies to treat metabolic diseases and prevent their devastating complications. We therefore formed a multidisciplinary research team to explore unique mechanisms that trigger macrophage activation.

The Notch pathway, one of the most fundamental cell signal transduction mechanisms, regulates embryonic development and differentiation of various cell types and organs (2). Such critical cell signaling pathways often play a role in normal adult homeostasis and disease processes in major organs. We therefore hypothesized that Notch signaling participates in disease mechanisms of cardiometabolic organs, including arteries, fat, and liver, and thus may serve as a common therapeutic target. We previously demonstrated that in cultured macrophages, Notch ligand Dll4 triggers various proinflammatory effects associated with atherosclerosis and metabolic disorders (1). To explore in vivo evidence, we used a clinically translatable study design with administration of anti-Dll4 blocking antibody to LDL receptor-deficient (Ldlr−/−) mice fed a high-fat, high-cholesterol diet, a commonly used model for atherosclerosis and metabolic disturbances in humans. Although pan-Notch inhibitors often cause severe toxicity, our Dll4 Ab did not have any obvious adverse effects and was well tolerated.

The Dll4 blockade attenuated atherosclerotic changes and macrophage accumulation in arteries in metabolically challenged Ldlr−/− mice, which may account for decreased activation of NF-xB, the key signaling pathway in inflammation, and its major target gene monocyte chemoattractant protein-1 (MCP-1), a potent chemokine that induces macrophage infiltration
(Fig. P1). Excessive accumulation of macrophages expressing collagen-degrading enzymes may cause disruption of coronary arteries and acute thrombosis (heart attack) (3). Dll4 blockade increased arterial collagen. Evidence also suggests that macrophages induce arterial calcification, a major health burden in aging societies (4). In the present study, Dll4 antibody administration reduced calcification. The same treatment also decreased MCP-1 expression and macrophage accumulation in adipose tissue (fat), which associated with reduced excessive fat deposition and decreased insulin resistance (Fig. P1). Notably, the present study used another model (leptin-deficient mice) of obesity and metabolic disorders to demonstrate similar improvements. We further found that Dll4 antibody treatment improved fatty liver without modulating the blood lipid profile.

Evidence suggests heterogeneity of macrophages and identified at least two subpopulations: proinflammatory “M1” macrophages and antiinflammatory/noninflammatory “M2” macrophages (5). Multiple studies have associated M1 macrophages with the metabolic syndrome and other inflammatory diseases. Our present study reveals that inhibition of Dll4-Notch reduces the inflammatory state of atherosclerotic arteries and fat, as gauged by decreased signs of M1 macrophages.

In the present study, we discovered that suppression of Dll4-Notch signaling attenuates several key components of cardiometabolic inflammation in parallel. We therefore propose that the Dll4-Notch axis participates in the shared mechanism for the cardiometabolic syndrome and is a unique target for much-needed therapies against this global health threat.