BRUTON’S TYROSINE KINASE INHIBITION TO SUPPRESS MAST CELL ACTIVATION IN Atherosclerotic Mice

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Aims
Acute cardiovascular diseases, such as myocardial infarction or stroke, are still a major cause of death in Western Society. The main underlying pathology of cardiovascular diseases is atherosclerosis, which is caused by the accumulation of lipids and inflammatory cells in the vessel wall, in so-called atherosclerotic plaques. Mast cells accumulate within these atherosclerotic plaques and activation of mast cells leads to the progression and destabilization of advanced plaques via the secretion of pro-inflammatory mediators and cytokines. Mast cells can be activated by various stimuli, of which crosslinking of the Fc receptor I (FcεRI) via IgE-antigen complexes is best known. Bruton’s tyrosine kinase (BTK), a cytoplasmic nonreceptor tyrosine kinase, is involved in the downstream signaling of FcεRI-mediated mast cell activation and degranulation. Therefore, BTK might be an attractive target to interfere in the FcεRI-mediated mast cell activation pathway. In this study, we thus aimed to assess the effects of the BTK inhibitor Acalabrutinib on FcεRI-mediated mast cell activation, plaque progression and destabilization in an atherosclerotic mouse model.

Methods and Results
Male LDLr−/− mice, 7–11 weeks old, were treated with Acalabrutinib (25 mg/kg p.o., n=15) or control solvent (n=14) three times per week for eight weeks. During treatment, mice were fed a Western-type diet (WTD) to induce atherosclerotic plaque formation. During the experiment, plasma total cholesterol levels and body weight did not differ between the control and treatment group. After eight weeks, mice were sacrificed and hearts were isolated to determine atherosclerotic plaque size and stability by histology. Other tissues, such as aorta, spleen and mediastinal lymph nodes were harvested to examine mast cell activation status and other immune cells by flow cytometry. After eight weeks of Acalabrutinib treatment in LDLr−/− mice, a significant 59% reduction in the frequency of CD117⁺ FcεRI⁺ mast cells was observed in aortic plaques of Acalabrutinib treated mice (0.24±0.06%) compared to control mice (0.57±0.08%, p<0.05), while relative mast cell activation status was not affected. Additionally, Acalabrutinib treatment inhibited B cell maturation in the circulation, spleen, mediastinal lymph nodes and peritoneal cavity of LDLr−/− mice compared to control mice. However, these effects on immune cells did not translate into effects on atherosclerosis, as Acalabrutinib treatment (size:12.3±2%; collagen:14.5±1.9%) did not significantly affect atherosclerotic plaque size and collagen content when compared to control mice (size:11.5±1.4%; collagen: 13.6±1.5%).

Conclusions
Conclusively, these findings suggest that Acalabrutinib treatment leads to reduced migration of mast cells to the atherosclerotic plaques of LDLr−/− mice, but does not directly affect mast cell activation and initial atherosclerotic lesion development.