Mast cell mediated neutrophil influx enhances plaque progression

I. Bot1; A. Wezel1; HM. Lagraauw1; D. Van Der Velden1; SCA. De Jager2; PHA. Quax3; J. Kuiper1
1Leiden Academic Centre for Drug Research, Leiden University, Biopharmaceutics, Leiden, Netherlands; 2University Medical Center Utrecht, Experimental Cardiology, Utrecht, Netherlands; 3Leiden University Medical Center, Surgery, Leiden, Netherlands
Rationale: Activated mast cells have been identified in atherosclerotic plaques and have previously been established to promote plaque progression and destabilization. As mast cells have the ability to release chemokines that mediate leucocyte fluxes, we propose that activated mast cells play a pivotal role in leucocyte recruitment during the development of atherosclerosis.

Methods and Results: Western-type diet fed B cell deficient apoE-/-muChain mice, which lack endogenous IgE, were systemically challenged with either IgE or PBS 6 times over a period of 8 weeks to induce mast cell activation during atherosclerotic lesion development. Mast cell activation in the aortic root was indeed significantly enhanced after IgE treatment (control: 35.2 ± 3.9% versus IgE: 48.2 ± 3.4% of activated mast cells, *P<0.05) and we observed a concomitant increase in plaque size (control: 2.0 ± 0.2*10E5 square mm versus IgE: 2.8 ± 0.3*10E5 square mm, *P<0.05). Intriguingly, a striking increase in the amount of perivascular neutrophils was observed in the IgE treated mice (control: 57.6 ± 10.6 neutrophils/square mm tissue versus IgE: 183.0 ± 38.7 neutrophils/square mm tissue; **P<0.05). In order to investigate whether activated mast cells can directly attract neutrophils, we injected C57Bl/6 or mast cell deficient KitW-sh/W-sh mice intraperitoneally with the mast cell activator compound 48/80. Mast cell activation led to a massive neutrophil influx into the peritoneal cavity (controls: 1.0 ± 0.6 versus compound 48/80: 5.1 ± 0.7, given in fold change, **P<0.01), while neutrophil numbers in mast cell deficient mice remained unaffected (controls: 1.0 ± 0.3 versus compound 48/80: 1.0 ± 0.2, P=NS). Furthermore, the newly recruited neutrophils were particularly CXCR2+ and/or CXCR4+. Interestingly, mast cells have been shown to secrete the CXCR2 and CXCR4 ligands CXCL1 and CXCL12, respectively. In vitro, supernatant of activated mast cells caused a 3-fold increase in neutrophil migration (32.3 ± 4.7*10E3 of migrated neutrophils versus 11.6 ± 2.5*10E3 neutrophils after stimulation with control supernatant, **P<0.01), which was seen to be inhibited by anti-CXCR2 (18.8 ± 2.2*10E3 neutrophils, *P<0.05), but not by the CXCR4 receptor antagonist AMD3100 (24.8 ± 6.9*10E3 neutrophils, P=NS).

Conclusions: In this study we demonstrate that chemokines, in particular CXCL1, released from activated perivascular mast cells induce neutrophil recruitment to the atherosclerotic plaque, thereby aggravating the inflammatory response which may further enhance atherosclerotic lesion progression and destabilization.