



Diagnosics of extracellular vesicles in acute myocardial infarction

Oxford supervisors: [Dr Naveed Akbar](#)¹, [Professor Robin Choudhury](#)¹

Department: 1. [Division of Cardiovascular Medicine, RDM](#)

Project Outline

Background: Diabetes is a negative predictor of outcomes for patients who have experienced an acute myocardial infarction (AMI), with higher rates of short and long-term mortality, reinfarction and heart failure. Diabetes drives exaggerated pro-inflammatory immune cell function (Edgar and Akbar et al. 2021) and endothelial cell activation, which increase myocardial infarct size and scar. However, it remains challenging to identify patients who would benefit from additional focused therapies in the immediate hours following presentation with AMI to hospital because we lack tools to identify these patients. We have shown that acutely after AMI extracellular vesicles (EV) are released into the peripheral blood and are enriched for the endothelial cell associated glycoprotein VCAM-1 on their surface and endothelial cell associated miRNAs within their membrane enclosed shells. These EV rapidly localise to the spleen and mobilise splenic-neutrophils, splenic-monocytes and induce their transcriptional activation prior to tissue recruitment. Importantly total plasma EV concentration correlates with the extent of myocardial injury, determined using oedema quantification by T2-weighted MRI, the scar size (determined by late gadolinium enhanced-(LGE)-MRI at 6 months post-AMI) and the degree of the peripheral blood neutrophil response in patients (Akbar et al. 2017 and 2022). Integrated protein and sphingolipid analysis (a predominant lipid group in EV membranes) of bulk plasma EV has shown diagnostic potential in AMI (Paget et al. 2022). Diabetes augments the number and composition of plasma EV in the blood of patients, therefore analysis of plasma VCAM-1⁺ EV may provide diagnostic utility in cardiometabolic disease.

Hypothesis: Early VCAM-1⁺ plasma extracellular vesicles are prognostic in diabetic patients following acute myocardial infarction.

Aims: (I) Integrated multi-omics analysis of plasma VCAM-1⁺ EV in a cohort of highly characterised AMI diabetic patients to generate mechanistically informative diagnostic tools. (II) Determination of the functional role of VCAM-1⁺ EV on innate immune cell function. (III) Generation of bioengineered VCAM-1⁺-EV to enable therapeutic immunomodulation for cardiometabolic disease.

Description: Early analysis of VCAM-1⁺ plasma EV may predict the immune response, injury size and subsets of patients for focused therapies. But until now it has not been possible to reliably isolate VCAM-1⁺ EV from the heterogeneous pool. I have developed the use of magnetic beads conjugated to anti-human-VCAM-1 antibodies to selectively isolate VCAM-1⁺-EV from the generalized plasma pool of circulating EV (Akbar et al. 2022). This immune-affinity platform allows high throughput sample processing for specific EV sub-population capture. Subsequently, captured EV can be released for analysis. We will extract the VCAM-1⁺ plasma EV at the time of presentation from control and diabetic

AMI patients versus a 6 month follow-up control (N=30 paired analysis per group, with additional recruitment if necessary) and determine the concentration of VCAM-1⁺ plasma EV, their whole transcriptome, protein, sphingolipid (80 sphingolipids) and metabolic cargo (150 metabolites). This study benefits from stored plasma samples and complementary patient specific cardiac MRI analysis of the heart in the acute phase (within 48 hours injury) and 6 months post-AMI (LGE), which is also complete and analyzed as part of the OxAMI study. This comprehensive analysis will determine, which VCAM-1⁺EC-EV constituent parts (EV concentration, protein, lipid, RNA and/or metabolic load) best predict the heterogeneity of the innate immune responses (i.e. leucocyte and lymphocyte counts) in the subsequent hours (6, 24 and 48 hours post-AMI) in diabetic and control AMI patients.

Expected Value of Results: We are limited by the ability to identify cardiometabolic patients who would benefit from additional therapies in the immediate hours post-AMI. The core principals of sub-population EV capture here, are easily extrapolated to a wide variety of cardiovascular and metabolic diseases with an inflammatory component.

Supervisor's recent relevant publications:

1. Paget et al. **Akbar**. Comparative and integrated analysis of plasma extracellular vesicle isolation methods in healthy volunteers and patients following myocardial infarction. 2022. *Journal of Extracellular Biology*.
2. **Akbar N.** et al. Rapid neutrophil mobilisation by VCAM-1+ endothelial extracellular vesicles. 2022. *Cardiovascular Research*.
3. Edgar L*, **Akbar N*** et al. *Joint First Authors. Hyperglycaemia induced trained immunity in macrophages and their precursors promotes atherosclerosis despite glucose normalisation. 2021. *Circulation*.
4. **Akbar N** et al. Endothelium-Derived Extracellular Vesicles Promote Splenic Monocyte Mobilization in Myocardial Infarction. 2017. *JCI Insight*.
5. Akawi N,...**Akbar N**, et al. Adipose Tissue Secretome Metabolomics Reveal C16:0-Ceramide as a Novel Endocrine Regulator of Vascular Redox State and Adverse Outcomes. 2021. *Journal of the American College of Cardiology*.