BAS/BSCR9 PROTEOMIC CHARACTERISATION OF EXTRACELLULAR SPACE COMPONENTS IN THE HUMAN AORTA

doi:10.1136/hrt.2010.205781.20

[1]A Didangelos, [1]X Yin, [2]K Mandal, [3]M Jahangiri, [1]M Mayr. *[1]King's BHF Centre, King's College London, UK; [2]Department of Cardiac Surgery, Johns Hopkins University, Baltimore, USA; [3]Department of Cardiac Surgery, St. George's Healthcare NHS Trust, London, UK*

Rationale Proteomics has been previously applied to vascular tissues, but few studies have specifically targeted the vascular extracellular matrix.

Methods and results In this study, we developed a novel methodology for the extraction and identification of extracellular proteins from human aortas. Our approach is based on (a) effective decellularisation to enrich for scarce extracellular proteins, (b) successful solubilisation and deglycosylation of matrix proteins and (c) relative estimation of protein abundance by liquid chromatography tandem mass spectrometry. This methodology resulted in the identification of 103 extracellular space proteins, of which one-third have never been reported in the proteomic literature of vascular tissues. In particular, our study revealed the presence of four novel glycoproteins in human aortas (podocan, sclerostin, agrin and asporin) and we confirmed their presence on the aortic extracellular matrix by independent methods. Although their function in the vasculature is currently unknown, we found that cholesterol loading regulated podocan and agrin expression in human aortic smooth muscle cells. Moreover, our methodology allowed us to investigate proteolytic activity within tissues, based on the identification of proteolytic enzymes and their corresponding degradation products. For instance, we were able to detect matrix metalloproteinase 9 by mass spectrometry and relate its presence to degradation of fibronectin.

Conclusions We expect this new proteomic method to shed light on the composition and breakdown of extracellular matrix within cardiovascular tissues and provide insights into important pathological processes, such as plaque rupture, aneurysm formation and restenosis.

BAS/BSCR10 DECORIN ACCELERATES VASCULAR CALCIFICATION VIA TRANSFORMING GROWTH FACTOR-β SIGNALLING MODULATION

doi:10.1136/hrt.2010.205781.21

G M Quigley, J Y Yan, S J Kakar, K Farrell, P A Kingston, C M Holt, M Y Alexander. *Cardiovascular Research Group, Faculty of Medicine & Human Science, School of Biomedicine, University of Manchester, UK*

Vascular calcification is a progressive pathology that occurs in many diseases, including atherosclerosis, diabetes, end-stage renal disease and valve disease. This study aimed to investigate whether decorin and transforming growth factor (TGF)-β signal modulation occurs during differentiation of vascular smooth muscle cells (SMCs). Decorin and osteopontin expression was identified in calcified human femoral arteries using immunohistochemistry and biochemical staining. A positive staining pattern of bone-related proteins and mineralisation was also found in aortic root sections from 8–16 week ApoE−/− mice fed a high-fat diet, particularly in the valve leaflets. These data suggest that this model, as well as being an established model of atherosclerosis, is also a suitable model to study vascular calcification. When adenoviral infection of human vascular SMC in vitro was used, decorin overexpression increased SMC osteogenic differentiation fourfold in comparison with controls, as assessed using alizarin red staining and alkaline phosphatase activity. The enhancement of mineralisation was reduced using two approaches to antagonise the TGF-β pathway—namely, adenoviral-mediated overexpression of the latency-associated particle of TGF-β- 1 (LAP-β1) and also a chemical inhibitor of TGF-β type I receptor, SB431542. In conclusion, these results suggest that vascular calcification involves the modulation of the decorin and TGF-β pathway in SMCs. The data may have implications for therapeutic targeting of this devastating pathology.

BAS/BSCR11 PULSE WAVE VELOCITY AS A SENSITIVE INDICATOR OF VASCULAR RISK ACROSS ETHNIC GROUPS: A EUROPEAN MALE AGEING (SUB-)STUDY (EMAS)

doi:10.1136/hrt.2010.205781.22

M Rezai, J Finn, F Wu, J K Cruickshank. *Department of Cardiovascular and Endocrine Sciences, University of Manchester, UK*

Introduction The incidence of vascular disease varies across different ethnic groups. People of South Asian (SA) origin have excess coronary disease (CHD) and diabetes, African-Caribbeans (AfC) less CHD despite more subjects having high blood pressure and diabetes than white Europeans (WE), which is poorly explained by standard risk factors. Having a simply measured intermediate outcome marker would be a considerable advance. Aortic pulse wave velocity (aPWV) is known to predict cardiovascular events and mortality but little is known of how it reflects risk across diverse ethnic groups.

Methods Community-based, volunteer AfC, SA and WE men aged 40–80 years were specifically invited from the larger numbers in the entire EMAS. Aortic pulse wave velocity (aPWV) was measured by arteriography.

Results AfC (n=61, age: 53±10 years) had a lower aPWV (7.42±1.5 m/s) than SA (n=56, age: 56±10 years, aPWV: 8.16±1.4 m/s) and WE (n=40, age: 58±8 years, aPWV: 8.06±1.1 m/s). This is despite higher systolic blood pressure (SBP) in AfC (129±16 mm Hg) than SA (123±14 mm Hg, p=0.026) and WE (125±12 mm Hg, p=0.1). the prevalence of diabetes was highest among the SA group (34%), followed by AfC (7%) and WE (0%). In multivariate analysis, aPWV in AfC was 0.69 m/s lower than SA (p=0.002), and 0.65 m/s than WE (p=0.006) adjusting for age, SBP, heart rate, diabetes status and body mass index.

Conclusion Differences in aPWV across ethnic groups here broadly reflect national patterns of cardiovascular morbidity/mortality. aPWV should be a useful target for intervention, in addition to SBP, across these groups.

Acknowledgements The research took place in, and was partly funded by, Wellcome Trust Clinical Research Facility, Manchester.

BAS/BSCR12 PROTEOMIC ANALYSIS OF SMOOTH MUSCLE CELLS DERIVED FROM CAROTID PLAQUE SHOWS DIFFERENCES BETWEEN SYMPTOMATIC AND ASYMPTOMATIC PLAQUES

doi:10.1136/hrt.2010.205781.23

[1]L E Full, [2]A H Davies, [1]R Wait, [1]C Monaco. *[1]Cytokine Biology of Atherosclerosis Group, Kennedy Institute of Rheumatology, Imperial College London, UK; [2]Imperial Vascular Unit, Imperial College London, Charing Cross Hospital London, UK*

Rationale Smooth muscle cells (SMC) are key players in atherosclerotic disease. The proteome of SMC in human carotid disease and its relationship with plaque-related symptomatology are still ill defined.

Methodology Carotid endarterectomy specimens were collected from 10 consenting patients (six symptomatic, four asymptomatic). Plaque