Purpose To investigate differences in plasma clot properties in patients with AF and CAD and compare the effect of warfarin and antiplatelets on clot structure in AF population.

Methods We studied 270 patients and divided them into 3 groups: AF on warfarin (n=184), AF on antiplatelets (n=46) and CAD (n=40). Plasma samples were obtained from participants and centrifuged to prepare platelet poor plasma. Assays were performed in 96-well polystyrene microtiter plates. Reagents were diluted in standard buffer (10 mM N-2-hydroxyethylpiperazine-N'-2-ethanesulphonic acid [HEPES], pH 7.4, 150 mM NaCl). Patient plasma samples (25 %) were incubated with tissue plasminogen activator (500 ng.mL-1) for 10 minutes at 37°C before the addition of CaCl2 (7.5 mM). Either PPF reagent (2.5 %), apPTT reagent (2.5 %), or thrombin (0.5 U.mL-1) were then added to initiate coagulation. Polymerisation of fibrin in plasma was monitored (AOD340 nm) using a Synergy H1 hybrid multi-mode plate reader, readings were taken in 12 second intervals for up to 60 minutes.

Results Comparisons between the 3 groups was performed using Kruskal-Wallis test, with Dunn post-hoc analysis and Holm-Sidak adjustment. There were no significant differences in clot structure between 3 subgroups. The maximum rate of clot formation was significantly delayed in the warfarin subgroup with all reagents used (p<0.001) (table 1). Plasma clot susceptibility to fibrinolysis increased with warfarin compared to antiplatelets but was significant only with APPT and thrombin reagents (p<0.001 and 0.04 respectively).

Conclusion Warfarin was effective in delaying clot formation compared to antiplatelets and also resulted in increased susceptibility of plasma clot to fibrinolysis.

Conflict of interest None

BS45 ACTIVATING TRANSCRIPTION FACTOR ATF2 NEGATIVELY REGULATES THE EXPRESSION OF ENDOTHELIAL NOTCH LIGANDS

Introduction ATF2 also known as cyclic AMP response element binding protein 2 (CREB2) is a member of the leucine zipper (bZIP) transcription factor family that binds to specific DNA sequences and regulates the transcriptional activation of target genes. ATF2 regulation and functions have been studied in a number of developmental and pathological conditions. Studies have shown that activation of ATF2 by VEGF mediates angiogenic processes such as endothelial cell migration and tubular morphogenesis but the molecular role of ATF2 in these processes is largely unknown. To shed some light on this matter we are identifying ATF2-target genes induced by stimulation of endothelial cells with pro-angiogenic stimuli.

Methods HUVEC were stimulated with various pro-angiogenic stimuli at different times, and the activation (phosphorylation) status of ATF2 was determined by Western Blot.

ATF2 functionality in endothelial cells was suppressed by infecting HUVEC with an adenovirus encoding a phosphorylation-mutant, dominant-negative version of ATF2 (Ad-ATF2AA) where phosphorylation residues Thr69 and Thr71 have been mutated to Ala. HUVEC infection with an adenovirus encoding GFP was used as a control. PCR-based screening of specific gene arrays was used to identify the effect of ATF2 loss-