P5378 Sildenafil affects tissue perfusion and the expression of proinflammatory molecules in a murine model of limb ischemia and atherosclerosis

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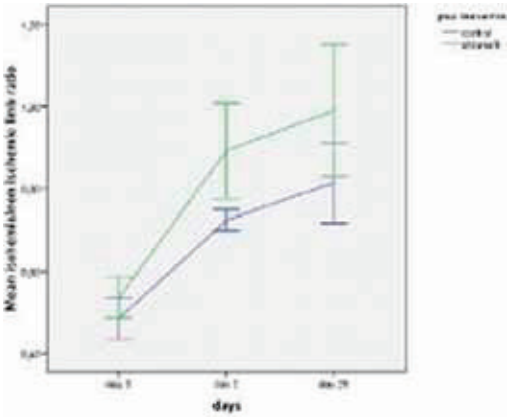
Purpose: Sildenafil, a phosphodiesterase type 5 antagonist has endothelium protective and angiogenic effects. We tested the hypothesis that sildenafil might improve tissue perfusion and neovascularization and downregulate proinflammatory molecules following limb ischemia.
Methods: ApoE-KO male mice, bred with cholesterol rich diet for 6 weeks, were anesthetized and underwent sunilateral hind-limb ischemia with ligation of the left femoral artery. Mice were randomized in three groups and received sildenafil (1mg/kg for 7 days in 0.4ml solution, intraperitoneally i.p.), or normal saline (0.4ml for 7days, i.p.). Bilateral hind-limb perfusion was estimated by laser Doppler perfusion imaging after surgery on days 0, 7 and 28. For capillary density assessment, the muscle tissue sections were stained with rat anti-CD31 antibody and assessed under confocal microscopy. sICAM-1, sE-Selectin and PAI-1 levels were evaluated at days 0 and 28 with enzyme-linked immunosorbent assay.
Results: Treatment with sildenafil was associated with significantly increased perfusion in the ischemic limb compared to control animals (Figure). The increase in blood flow was maintained at day 28. Ischemia exerted no significant effects on sICAM (from 1.91±0.15 to 0.140±0.19 ng/ml, p=NS), sE-Selectin (from 4.23±0.877 to 2.69±1.00 ng/ml, p=NS) and PAI-1 levels (from 0.219±0.055 to 0.126±0.054 ng/ml, p=NS). Sildenafil significantly decreased sICAM-1 (from 2.1±0.15 to 1.2±0.12 ng/ml, p<0.01), sE-selectin (from 5.34±0.4 to 2.45±0.51 ng/ml, p<0.01) and PAI-1 (from 0.13±0.02 to 0.07±0.012 ng/ml) levels.



Conclusions: Sildenafil exerts significant beneficial effects on tissue perfusion and neovascularisation after limb ischemia and downregulates adhesion molecules in the atherosclerotic milieu.

P5379 Abnormal redox state of protein disulfide isomerase in the diabetic heart

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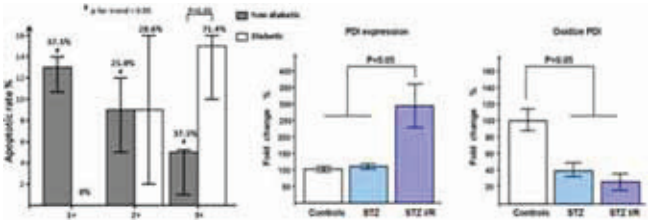
Purpose: Diabetes is associated with an increased risk of heart failure, in part explained by increased apoptosis. Endoplasmic reticulum stress has been suggested as a mechanism of increased apoptosis. Protein disulfide isomerase (PDI) is a member of the unfolded protein response that prevents apoptosis in cardiomyocytes under stress. We hypothesized that diabetes impairs PDI expression or function by an alteration in its redox state.
Methods: Surgical myocardial biopsies harvested from the antero-lateral left ventricular wall at time of coronary artery bypass surgery in diabetic (N=7) and non-diabetic (N=8) patients, were used to assess PDI expression (1 to 3 scale-immunohistochemistry) and cardiomyocytes death (TUNEL). A mouse model of diabetes, induced by streptozotocin (STZ) injection (130 mg/ml), was used to study PDI redox state and expression after ischemia reperfusion (I/R) injury induced by 30 minutes occlusion of the left anterior descending coronary artery followed by 7 days reperfusion.
Results: Diabetic patients had significantly greater apoptosis than non-diabetic patients (P=0.05), and tended to have greater myocardial PDI expression (P=0.11). PDI expression was inversely correlated with apoptosis in the non-diabetic patients (R=-0.89, P<0.01), but not in the diabetic patients. PDI expression in diabetic mice showed an increase in the total expression after I/R, but diabetic state was characterized by a reduction of the oxidized (catalytically active) form of PDI, and a worse cardiac remodeling following I/R.



Conclusions: An increase in PDI levels with a paradoxical decrease of its catalytically active form occurs in the diabetic heart after ischemia and explains the lack of protective effects of PDI in diabetes.

P5380 Novel technology enhances specificity of exercise testing in women referred for angiography

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Background: Exercise testing (ETT) in women with suspected coronary artery disease (CAD) has limited specificity, leading to an excessive rate of unnecessary angiographies. Analysis of high-frequency QRS (HFQRS) components was recently shown to be more accurate than ST changes in identifying stress-induced ischemia, independent of gender. We aimed to evaluate the diagnostic value of HFQRS in women referred to angiography.
Methods: Analysis was performed in 72 women (age 64±10 yo) referred to non urgent angiography, who were able to perform ETT and attained at least 70% of their maximal age-predicted heart rate. ETT was done using the HyperQ system, which provides both conventional ECG and HFQRS signals. HFQRS diagnosis was determined by computerized analysis, measuring the stress-induced reduction in HFQRS intensity. The diagnostic performance of HFQRS, computerized ST segment analysis and clinical interpretation of the ETT were compared, using angiography as gold standard. Significant CAD was defined as stenosis of 70% in a single vessel, or 50% in the left-main artery.
Results: HFQRS analysis was available in 54 pts. Of these patients, 18 (33%) had significant CAD and 36 pts (66%) had insignificant or no stenosis. ETT was considered abnormal in 35 pts. Both ETT interpretation and ST analysis had low specificity (Table). HFQRS analysis provided significantly higher specificity of 86% (P<0.01 vs ETT or ST changes), with comparable sensitivity of 78%.

	Sensitivity (N=18)	Specificity (N=36)	Accuracy (N=54)
HFQRS	78%	86%*	83%*
Computerized ST	72%	50%	57%
ETT interpretation	89%	47%	61%

*P<0.02 vs. ETT or ST changes.

Conclusions: HFQRS analysis improved the specificity and the overall accuracy of ETT in diagnosing CAD, while retaining high sensitivity. Thus, HFQRS may reduce the number of unnecessary angiography procedures in women.

P5381 Association between -374T/A and -429 T/C polymorphisms of the RAGE gene and myocardial infarction

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Background: Inflammation and genetics play an important role in the pathogenesis of coronary artery disease (CAD). The role of genetics in CAD and myocardial infarction (MI) pathogenesis have not been extensively explored. Functional polymorphisms in the receptor for the advanced glycation end products (RAGE) gene have been linked to RAGE expression and play a crucial role in modulating the inflammatory response. Previous studies have reported an association between the homozygous AA genotype of the -374 T/A polymorphism and reduced risk for CAD at angiography.
Purpose: Aim of the present study was to identify possible relationship between two common functional polymorphisms (-429 T/C and -374 T/A) in the promoter region of the RAGE gene and MI in a consecutive cohort of Caucasian patients.
Methods: a total of 691 patients with incident first MI and 180 age-matched white individuals were investigated. Genotyping of the –374T/A and -429 T/C RAGE polymorphisms were performed by means of PCR-RFLPs. To compare the age at onset of MI across the RAGE genotypes Kaplan–Meier curves were computed.
Results: The genotype and the allele distribution of the -429 T/C polymorphism was not statistically different in MI patients compared to controls.
The AA genotype frequency of the -374 T/A polymorphism was significantly lower in patients with MI than controls (13.4% vs 22.7% p< 0.01). The MI patients showed lower frequency of A allele compared to controls (37% vs 45% p<0.01). After adjustment for potential confounders (other well-established risk factors)