P-432
SYNERGISTIC EFFECT BETWEEN ENOS GENE G894T AND GNB3 GENE C825T POLYMORPHISMS IN PATIENTS WITH ESSENTIAL HYPERTENSION
Dongbao Li, Qi Hua, Lin Pi. Cardiology, Beijing Xuanwu Hospital, Beijing, China.

Aim: To investigate an interaction between the G894T polymorphism of endothelial nitric oxide synthase (eNOS) gene and the C825T polymorphism of GNB3 in the risk for essential hypertension (EH).

Methods: Polymerase chain reaction combined with restriction enzyme digestion was used to detect the G894T polymorphism of eNOS gene and C825T polymorphism of GNB3 gene in 151 normotensive controls and 310 hypertensive patients.

Results: The frequencies of T allele and GT+TT genotype of eNOS gene G894T polymorphism in EH were significantly higher than in control group; There were no significant difference of the GNB3 gene C825T polymorphism CC,CT,TT genotypes and C,T alleles between hypertensive patients and normotensive controls; The odds ratio (OR) estimated by combined analysis with eNOS 894T and GNB3 825T (1.82) was markedly increased compared with that estimated alone from either eNOS 894T allele (0.504) or GNB3 825T allele (0.954).

Conclusions: ENOS 894T allele should be a risk factor for EH in Chinese Han nationality, there was a significant synergistic effects of eNOS 894T allele and GNB3 825T allele in EH.

Key Words: Essential Hypertension, Polymorphism, Synergistic Effects

P-433
SYNERGISTIC EFFECT BETWEEN ENDOTHELIAL NITRIC OXIDE SYNTHASE GENE T786C AND 4B/A POLYMORPHISMS IN PATIENTS WITH ESSENTIAL HYPERTENSION
Dongbao Li, Qi Hua, Lin Pi. Cardiology, Beijing Xuanwu Hospital, Beijing, China.

Aim: To investigate an interaction between eNOS gene T786C and 4b/a polymorphisms in the risk for essential hypertension (EH).

Methods: Polymerase chain reaction (PCR) combined with restriction enzyme digestion was used to detect the T786C and 4b/a polymorphisms of eNOS gene in 189 normotensive controls and 310 hypertensive patients.

Results: The frequencies of a allele, ab+aa genotype and C allele, CC+CT genotype in EH were significantly higher than in control group respectively; The odds ratio (OR) estimated by combined analysis with eNOS 786C and a allele (2.51) was markedly increased compared with that estimated alone from either eNOS 786C (0.542) or a allele (0.389).

Conclusions: ENOS 786C and a allele should be a risk factor for EH in Chinese Han nationality; there was a significant synergistic effects of eNOS 786C and a allele in EH.

Key Words: Essential Hypertension, Gene Polymorphism, Nitric Oxide Synthase (eNOS)

P-434
AMINOPEPTIDASE B-INHIBITION ATTENUATES COLLAGEN PRODUCTION IN CARDIAC FIBROBLASTS THROUGH INHIBITION OF LYSYL OXIDASE

The aim of this study was to determine whether the aminopeptidase inhibitor arphamenine A could affect the collagen production and expression in control and TGF-beta1-treated cardiac fibroblasts. Cardiac fibroblasts from passage 2 from normal male adult rats (n = 5) were cultured to confluence, incubated with (out) 600 pmol/l TGF-beta1 for 2 days in serum-free Dulbecco’s modified Eagles medium and then incubated with 100 pmol/l arphamenine A for 1 day in this medium with added ascorbic acid, beta-aminopropionitrile and titrated proline. Soluble collagen was measured in the conditioned medium and nonsoluble collagen in the cell layer. Aminopeptidase activity was estimated by spectrophotometric determination of the liberation of p-nitroaniline from alanine and arginine-p-nitroanilide. Matrix metalloproteinase (MMP) and lysyl oxidase activity were assayed in the conditioned medium. A semi-quantitative reverse transcription polymerase chain reaction was used to determine the expression of lyses oxidase and collagen type I and III. Arphamenine A dose-dependently inhibited the basal and TGF-beta1-stimulated aminopeptidase activity. Arphamenine A reduced the soluble and nonsoluble collagen production in control and TGF-beta1-treated cardiac fibroblasts, while it decreased the collagen type I and III expression only in TGF-beta1-fibroblasts. Lysyl oxidase, MMP-1 and MMP-2 activity were inhibited by arphamenine A in the conditioned media of control and TGF-beta1-treated cardiac fibroblasts, while MMP-13 and TIMP-2 were not affected by arphamenine A. Our data show that the specific aminopeptidase inhibitor arphamenine A reduces the collagen production in cardiac fibroblasts and that this reduction is accompanied by a pronounced inhibition of lyses oxidase.

Key Words: Cardiac Fibroblasts, Collagen, Transforming Growth Factor-Beta

P-435
INHIBITION OF COLLAGEN GEL CONTRACTION IN CARDIAC FIBROBLASTS BY IMPRIMARE

The aim of this study was to determine whether (amino)peptidase inhibition could affect the collagen gel contraction in basal (control) cardiac fibroblasts. Cardiac fibroblasts (from 6 normal male adult rats) were cultured to confluence in Dulbecco’s modified Eagles medium (DMEM) with 10% fetal bovine serum (FBS). These fibroblasts (100,000 cells) were then further incubated in a floating collagen gel lattice with various concentrations (1, 10, 50, 100 or 300 μmol/l) of imipramine, a puromycin-sensitive aminopeptidase inhibitor, or with des-Tyr-Met-epinephrine (10 or 100 μmol/l), a neutral endopeptidase (NEP-24.11) or enkephalase inhibitor, and compared with the NEP inhibitors phosphoramidon or thiorphan (10 μmol/l) for 3 days in DMEM without FBS. The contraction of the collagen gel lattice by cardiac fibroblasts was determined by measuring the gel volume with titrated water. The original gel volume of the hydrated collagen gel in the absence of fibroblasts averaged 240.1 ± 3.2 μl and decreased to 148.1 ± 9.7 μl in control fibroblasts. Imipramine at a concentration of 1 μmol/l did not affect the gel volume of control fibroblasts (146.7 ± 9.6 μl). The gel volume is however increased with higher concentrations of imipramine to , respectively ,164.7 ± 9.6, 222.0 ± 12.9, 238.3 ± 1.4 and 243.8 ± 5.4 μl with 10, 50, 100 and 300 μmol/l. The latter 2 concentrations of imipramine even completely inhibited the collagen gel contraction in fibroblasts and this