AMYLOID BETA 1-40 AND ITS UPSTREAM REGULATORY PATHWAY BACE1-AS LONG NONCODING RNA/BACE1 ARE ASSOCIATED WITH PRESENCE AND SEVERITY OF HUMAN ATHEROSCLEROTIC DISEASE

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Background

- Long noncoding RNAs (lncRNAs) are a group of non-protein-coding RNAs > 200 nucleotides and participate in biological processes and pathophysiological conditions in vivo or in vitro. Recently, more and more lncRNAs interfering with the progress of atherosclerosis are identified and characterized in the atherogenic cells such as vascular smooth muscle cells (VSMCs), endothelial cells (ECs) and monocytes/macrophages showing that lncRNAs play an important role in the occurrence of atherosclerosis.
• β-Secretase 1 (BACE1) is implicated in Alzheimer’s disease as the enzyme responsible for the rate-limiting step in β-amyloid (Aβ) production, but may also be involved in a wide range of cardiovascular disorders by regulating β-amyloid (Aβ) production.

• The most common b-amyloid isoforms are b-amyloid 1-40 (Abeta40) and 1-42, typically produced by division that occurs in the endoplasmic reticulum or in the trans-Golgi network.

• Abeta40 activates a cascade of proinflammatory events in endothelial cells and macrophages including cytokine secretion and oxidative stress that lead to vascular disease.

• Abeta40 is the main amyloid-beta peptide found in human atherosclerotic lesions.

• Abeta40 levels are significantly and independently associated with arterial stiffness progression, incident subclinical atherosclerosis and incident coronary heart disease.

• Antisense (AS) lncRNAs are transcripts emerging from the opposite strand of a coding-RNA region.
• BACE1-AS is transcribed from an intron of the b-secretase-1 (BACE1) gene in antisense direction, completely overlapping exon 6.

• BACE1-AS enhances the stability of the BACE1 sense transcript.

• BACE1-AS has been shown to enhance BACE1 stability and might amplify its pro-atherosclerotic role. However, the in vivo interplay between BACE1/BACE1-AS and cardiovascular indices has not been studied in a clinical setting of graded atherosclerosis severity, ranging from subclinical to overt disease.

Objectives

- To evaluate associations of BACE1/BACE1-AS levels in human peripheral blood mononuclear cells (PMBCs) with established markers of increased cardiovascular risk and atherosclerosis burden in individuals with and without clinically overt cardiovascular disease (CVD).
Methods

➢ BACE1/BACE1-AS expression was measured in samples of PMBCs derived from 43 patients with stable coronary artery disease (CAD), 26 subjects with acute myocardial infarction (AMI) and 145 individuals without evidence of CVD.

➢ Structural and functional vascular measurements were used as surrogate markers of subclinical CVD:

1. **PWA (Pulse wave analysis)**

   • PWA allows non-invasive assessment of aortic hemodynamics
   • Central aortic BP is superior predictor of CV risk over peripheral BP
   • Augmentation index (AI) indicates arterial wave reflections
   • Arterial wave reflections are major determinants of SBP
   • AI correlates with CV mortality

Vlachopoulos et al, EHJ 2010
Arterial tonometry
2. **PWV (pulse wave velocity)**

- Increased PWV
- Reflects increased aortic stiffness
- Aortic stiffness is a major determinant of SBP
- Indicates target organ damage
- Predicts future adverse CV events

**Simultaneous recording**
Carotid and Femoral artery sites

\[ PWV = \frac{D}{T} \]

*Vlachopoulos C et al. JACC 2010*
*Ben-Shlomo, et al. JACC 2013*
*Vlachopoulos C, et al. JACC 2013*
3. Ankle-Brachial Index (ABI)

- Low ABI is a non-invasive marker of:
  1. Flow-limiting peripheral arterial disease
  2. A marker of CV mortality

Hiatt, WR. N Engl J Med 2001; 344:1608
4. **FMD (Flow Mediated Dilatation)**

1. Baseline measurement
2. Ischemic occlusion (5mins)
3. Occlusion release $\Rightarrow$ Reactive hyperemia + Increase in diameter

Automatic detection of lumen diameter changes

For every 1% increase in FMD, 8% decrease in CVD incidence
5. Intima-Media Thickness (IMT)

NORMAL IMT

IMT THICKENING

NON-OBSTRUCTIVE PLAQUE (IMT > 1.5 mm)

OBSTRUCTIVE PLAQUE

Lorenz M, Circulation, 2007
• The number of angiographically confirmed diseased coronary arteries was used to assess the extent of CAD in patients with established CVD.

• Cardiovascular risk factors (CVRFs), including impaired glomerular filtration rate below 60 ml/min, smoking, hypertension, hyperlipidemia, diabetes and obesity were recorded for each participant. C-reactive protein (CRP) was measured as an inflammatory marker.

• Standard cardiac ultrasound assessment was performed, including measurement of left ventricular ejection fraction (LVEF) by Simpson’s biplane method.

• Exclusion criteria for the stable CAD and the AMI groups included autoimmune disease, cancer, acute renal failure, acute stroke, chronic inflammatory disease or active infection.
Results

- BACE1 and BACE1-AS levels escalated across increasing (i.e. no overt CVD-stable CAD-AMI) severity of atherosclerosis (p for trend<0.001 for both).

- BACE1-AS but not BACE1 levels were marginally increased in AMI patients as compared to stable CAD individuals (p=0.06).

- Within the non-CAD group, BACE1 (p=0.031) and BACE1-AS (p=0.006) were substantially increased in individuals with aggregated CVRFs in comparison to participants of low CV risk.
Among CAD patients, BACE1-AS was associated with increased CRP (p=0.029) and impaired LVEF below 50% (adjusted OR=1.92 per 1-SD increase, 95% CIs 1.01-366, p=0.047) after controlling for the effect of age, gender and CVRFs.

BACE1-AS was related to decreased time to return of the reflected arterial waves (adjusted coefficient=-0.036ms per 1-SD increase, p=0.036).

CAD patients distributed in the highest tertile of BACE1 levels presented a 5-fold increase in the odds for multi-vessel disease (OR=5.1 for highest versus lower tertile of BACE1, 95% CI 1.67-15.7, p=0.004).
Conclusions

➢ BACE1 and BACE1-AS levels in human PMBCs correlate with the severity of atherosclerosis and are associated with increased disease burden in patients with established CAD.

➢ BACE1 and BACE1-AS might constitute higher-order regulators of detrimental patho-mechanisms and may serve as potential upstream therapeutic targets in respect to CV precision-medicine.

➢ Further studies are warranted to replicate our findings and elucidate their clinical implications in risk stratification of patients with subclinical and overt atherosclerosis.
Thank you