Increased Dipeptidyl Peptidase-4 Accelerates Diet-Related Vascular Aging and Atherosclerosis in ApoE-Deficient Mice under Chronic Stress

Department of ICU
Yanbian University Hospital

Yanna Lei MD, PhD;
Xianwu Cheng, MD, PhD, FAH

December 9, 2017
Stress and Disease

- Inflammation
- Mental disease
- Metabolic syndrome
- Hematopoietic stem cell activation
- Cardiovascular disease
DPP-4 and its inhibitors

DPP-4 is a complex enzyme that acts as a membrane-anchored cell surface exopeptidase that truncates a large number of peptides (e.g., hormones, cytokines, and growth factors). DPP-4 has gained considerable interest as a therapeutic target, and a variety of DPP-4 inhibitors that prolong the insulinotrophic effects of glucagon-like peptide-1 (GLP-1) are widely used in clinical settings as antidiabetic drugs.
DPP-4 and its substrates

**Signaling-related peptides**
- SP
- SDF-1 α/β
- GM/G-CSF
- PYY
- GIP
- GLP-1
- GLP-2

**Chemokines/cytokines**
- CXCL2 (Gro β)
- CXCL6 (GCP2)
- CXCL9 (MIG)
- CXCL10 (IP10)
- CXCL11 (ITAC)
- IL-2
- IL-3
- MCP-1
- MCP-3
- HMGB1
- IL-1β
- Bradykinin
- MCP-2
- GCP-2
- TPO

**Hormones/peptides**
- GHRF
- IGF
- APN
- Prolactin
- EPO
- PYY
- BNP

**Neuropeptides**
- NPY

**Other peptides**
- Adprogin
- CG
- Enterostatin
- Endomorphin-2
- β-casomorphin-2
- CLIP
- Tyr-melanostatin
- α 1-microglobulin

Chronic psychological stress increased blood DPP-4 levels in a time dependent manner.

Zhu, Lei and Cheng. JAHA 2017;6:e006439
CAD and ACS patients had increased levels of plasma DPP-4
The aim of our study was to investigate the effects of DPP-4 inhibitor on vascular aging and atherosclerotic plaque growth and the related mechanisms with special focusing on APN-PPAR α signaling activation in ApoE-/- mice under chronic psychological stress.

APN: adiopoectin; PPAR-α: Peroxisome Proliferator-Activated Receptor;
Sampling Protocol (1)

Birth

6-week-old male ApoE\(^{-/-}\) mice (KOR/StmSlc background)

6 weeks

18 weeks

High fat diet

Group 1: HFD (Control)
Group 2: HFD + Stress (HD-S)

Sampling

Blood
Aortas
Effects of stress on plasma lipid profile and DPP4, leptin, GLP-1, and APN levels

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Non-stress</th>
<th>Stress</th>
</tr>
</thead>
<tbody>
<tr>
<td>T-ch (mg/dL)</td>
<td>575.2 ± 14.3</td>
<td>562.5 ± 15.8</td>
</tr>
<tr>
<td>HDL-C (mg/dL)</td>
<td>23.1 ± 2.2</td>
<td>23.0 ± 2.3</td>
</tr>
<tr>
<td>Triglyceride (mg/dL)</td>
<td>135.1 ± 4.8</td>
<td>73.5 ± 4.0**</td>
</tr>
<tr>
<td>NEFA (µEQ/L)</td>
<td>194 ± 12</td>
<td>106 ± 12**</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>3.9 ± 0.3</td>
<td>4.2 ± 0.3</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>0.5 ± 0.0</td>
<td>0.8 ± 0.0</td>
</tr>
<tr>
<td>Glucose (mg/dL)</td>
<td>39.3 ± 2.6</td>
<td>36.1 ± 2.9</td>
</tr>
<tr>
<td>DPP4 (ng/L)</td>
<td>305 ± 28</td>
<td>823 ± 34**</td>
</tr>
<tr>
<td>Leptin (pg/ml)</td>
<td>402 ± 45</td>
<td>177 ± 25**</td>
</tr>
<tr>
<td>GLP-1 (pM)</td>
<td>15.9 ± 1.1</td>
<td>9.2 ± 0.8**</td>
</tr>
<tr>
<td>APN (ng/mL)</td>
<td>7577 ± 382</td>
<td>5619 ± 598*</td>
</tr>
</tbody>
</table>

T-ch: total cholesterol; HDL-C: high-density lipoprotein cholesterol; NEFA: nonesterified fatty acid; BUN: blood urine nitrogen; DPP4: dipeptidyl peptidase-4; GLP-1, glucagon like protein-1; APN, adiponectin. Data are mean ± SEM. * $P<0.05$, ** $P<0.01$ by ANOVA and Tukey’s post hoc tests.
Stress reduced subcutaneous/inguinal adipose and body weight (BW)
Stress accelerated vascular senescence and plaque lipid accumulation and growth
Stress reduced plaque collagen volume and promoted elastin degradation.
Stress enhanced mac infiltration, inflammatory chemokine expression and neovessel formation.
Stressed aortas had increased levels of MMP-2/-9, TIMP1/2, CatS/K/Land APN and decreased eNOs genes.
Stress increased levels of AT1R and gp91phox and decreased levels PPAR-α, except p-AMPK proteins.
Protocol (2)

Sampling
Blood
Aortas

Group 1: HFD+Stress (Control)
Group 2: HFD+Stress+Anagliptin (S-Ana)
DPP4 inhibition increased levels of APN and GLP-1 proteins

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Stress</th>
<th>S-Ana</th>
</tr>
</thead>
<tbody>
<tr>
<td>Triglyceride (mg/dL)</td>
<td>21.1 ± 2.2</td>
<td>13.6 ± 3.1*</td>
</tr>
<tr>
<td>LDL (mg/dL)</td>
<td>47.6 ± 6.1</td>
<td>38.1 ± 12.5</td>
</tr>
<tr>
<td>HDL (mg/dL)</td>
<td>4.3 ± 0.2</td>
<td>5.0 ± 0.0</td>
</tr>
<tr>
<td>NEFA (μEQ/L)</td>
<td>152 ± 8</td>
<td>136 ± 13</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>3.2 ± 0.4</td>
<td>4.1 ± 0.8</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>0.5 ± 0.0</td>
<td>0.5 ± 0.0</td>
</tr>
<tr>
<td>DPP4 (ng/ L)</td>
<td>976 ± 4</td>
<td>477 ± 22**</td>
</tr>
<tr>
<td>Leptin (pg/ml)</td>
<td>214 ± 9</td>
<td>301 ± 25**</td>
</tr>
<tr>
<td>GLP-1 (pM)</td>
<td>11.3 ± 0.6</td>
<td>19.4 ± 0.8**</td>
</tr>
<tr>
<td>APN (ng/mL)</td>
<td>5574 ± 417</td>
<td>8492 ± 584**</td>
</tr>
</tbody>
</table>
DPP4 inhibition mitigated vascular aging and plaque growth and collagen/elastin metabolism
Anagliptin inhibited macrophage infiltration and inflammatory chemokine expression
DPP4 inhibition mitigates the changes in AT1R, PPAR-α, p-AMPK, and gp91phox proteins
DPP4 inhibition reduced MMP-2/-9 activity
GLP-1 analogues exenatide stimulated APN expression in adipocytes in a dose-dependent manner, but not by anagliptin.
GLP-1 analogues exenatide improved atherosclerotic lesion formation

Oil red O staining

Yang and Lei et al. Atherosclerosis 2017;264:1-10
Proposed mechanisms

Chronic Stress

- Upregulates DPP4
- Downregulates GLP-1

APN

Oxidative stress:
- gp91phox
- p47phox
- P67phox
- Superoxide production

Bone marrow

Proteolysis:
- CatS/K
- MMP-2/-9

HSC activation

Vascular aging
Atherosclerotic plaque growth and instability

Inflammation

Conclusion

These results indicate that the DPP-4 inhibition-mediated benefits are likely attributable, at least in part, to attenuation the plaque inflammation, oxidative stress and proteolysis associated with GLP-1-mediated APN production in ApoE\(^{-/-}\) mice under stress. Thus, DPP-4 will be a novel therapeutic target for the treatment of stress-related cardiovascular disease.
Thank You!

谢谢