Novel Insights into the Production and Atherogenicity of Lipoprotein(a)

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Outline

• Current understanding of how Lp(a) concentrations are specified

• Recent breakthroughs in our understanding of Lp(a) as a risk factor for CHD

• Novel insights into the pathogenic mechanisms of Lp(a)
Structure of Lipoprotein(a) [Lp(a)]

apolipoprotein(a) [apo(a)]

low-density lipoprotein (LDL)

Boffa MB, in Therapeutic Lipidology (2008)
Comparison of Apo(a) and Plasminogen

Lipoprotein(a) [Lp(a)] Levels

- Plasma Lp(a) levels vary over 1000-fold in the population from <1 to >100 mg/dL
  - Risk threshold: 75th percentile; ethnic group-specific

- A general inverse correlation has been observed between apo(a) isoform sizes and plasma Lp(a) levels

- Lp(a) levels are primarily genetically determined:
  - Greater than 60% of observed variability from LPA itself
    - number of KIV₂ repeats
    - pentanucleotide [(TTTTA)ₙ] repeat in promoter
    - rs3798220 (Ile4399Met); rs10455872 (intronic)
  - Remainder: polygenic and non-genetic factors?
  - Lp(a) levels resistant to conventional methods for LDL lowering

- Lp(a) levels are determined primarily at the level of production rather than catabolism
Control of Lp(a) Production

assembly

folding & secretion

transcription

(TTTA)$_n$

(KIV)$_n$

rs10455872 (intrinsic)

rs3798220 (Ile4399Met)
Lp(a) as a Risk Factor for CHD

- **Retrospective Case-Control Studies**
  - Numerous studies all show that Lp(a) is elevated in Cases versus Controls

- **Prospective Studies**
  - Although results are less consistent, several meta-analyses show that Lp(a) is a risk factor for CHD
    - Erqou et al., Emerging Risk Factors Colloquium. *JAMA* 2009;302:412
  - Some evidence to suggest that Lp(a) risk may be dependent on elevated LDL levels although results are inconsistent

- **Summary:** Research is consistent with Lp(a) as an independent risk factor that shows continuous association with CHD, particularly at extreme high concentrations of Lp(a)
Validation of Lp(a) as a Treatment Target: Key Results from Human Genetic Studies

Three-stage genome-wide haplotype association study

- Trégouet et al., *Nature Genet.* 2009;41:283
  - Gene cluster containing *LPA* implicated in CHD risk
  - 2 *LPA* haplotypes were identified as highly significant for CAD

Mendelian randomization study

- Kamstrup et al., *JAMA.* 2009;301:2331
  - First study of its type
  - Measured genetic variation in *LPA*: kringle number polymorphism using real-time PCR

Candidate gene array identified associations between 2 *LPA* variants and CAD

- Clarke et al., *NEJM* 2009;361:2518
  - Variants are rs10455872 (OR 1.7) and rs3798220 (OR 1.92)
  - Accounted for 36% of variation in Lp(a) levels
  - Relationship abolished after adjusting for levels
Genetically Elevated Lp(a) and Risk of MI
Mendelian Randomization Study

MI risk by quartiles of Lp(a)

<table>
<thead>
<tr>
<th>Percentile</th>
<th>mg/dL</th>
<th>Participants, No.</th>
<th>Events, No.</th>
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<td>&gt;95th</td>
<td>&gt;117</td>
<td>376</td>
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<tr>
<td>90th-95th</td>
<td>77-117</td>
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<tr>
<td>67th-89th</td>
<td>30-76</td>
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<td>5-29</td>
<td>3385</td>
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<td>&lt;22nd [Reference]</td>
<td>&lt;5</td>
<td>1582</td>
<td>104</td>
</tr>
</tbody>
</table>

Lp(a) level by octiles of KIV-2 repeats

Octiles of KIV-2 repeats

P < .001

MI risk by quartiles of KIV-2 repeats

Kamstrup et al., JAMA 2009;301:2331-2339
Association *LPA* Genotype Score with Lp(a) Level and CHD Risk – PROCARDIS Cohort

Clarke et al. *NEJM* 2009;361:2518
Pathogenic Roles of Lp(a) in the Vessel Wall

Multiplicity of Proposed Pathogenic Mechanisms for Lp(a)

- ↑ Foam cell formation
- ↑ Oxidized PL
- ↑ Monocyte chemoattractant activity
- ↑ SMC proliferation
- ↑ EC dysfunction

- ↓ Plasminogen activation
  (platelets/ECs/fibrin/degraded fibrin)
- ↓ TFPI activity
- ↓ Clot permeability
- ↑ EC PAI-1 expression
- ↑ platelet responsiveness

Modified from Koschinsky & Marcovina Curr. Opin. Lipidol. 2004;15:167
Role of OxPL in Lp(a) Function

- Atherogenicity of Lp(a) may be mediated in part by its association with proinflammatory OxPL (present on apo(a) and LDL)
  - Tsimikas et al., NEJM 2005;353:46

- In plasma, OxPL preferentially associate with Lp(a)
  - Bergmark et al., JLR 2008;49:2230

- Extent of Lp(a) OxPL modification is inversely correlated with apo(a) isoform size
  - Tsimikas et al., Circulation 2009;119:1711

- EPIC-Norfolk study: additive (and possibly independent) effects of elevated Lp(a) and OxPL
  - Tsimikas et al., JACC 2010;56:946
Role of OxPL in Lp(a) Function

Tsimikas JACC. 2006;47:2219

Keichl ATVB. 2007;271788

Tsimikas JACC. 2010;56:946
Site of Oxidized Phospholipid Addition in Apo(a)
Site of Oxidized Phospholipid Addition in Apo(a)
Role of OxPL in Pro-inflammatory Effects of Apo(a)
Role of OxPL in Stimulation of Macrophage Apoptosis by Apo(a)

- OxPL on apo(a) trigger apoptosis in ER-stressed macrophages (Seimon T, Cell Metab 2010;12:467)
- Trypsin-treated 17K retained effect; PLA₂/trypsin-treated 17K lost effect
- Thus, OxPL is active moiety

(Seimon T, Cell Metab 2010;12:467)
Role of OxPL in Stimulation of Macrophage Apoptosis by Apo(a)

(Seimon T, Cell Metab 2010;12:467)
Link Between Endothelial Function and Vascular Disease

Normal endothelium
- Anticoagulant
- Anti-inflammatory
- Impermeable

Damaged endothelium
- Procoagulant
- Pro-inflammatory
- Permeable

- NO
- TM
- tPA
- PGI2
- Heparin-like proteoglycans
- E-selectin
- ET-1
- TF
- PAI-1
- VCAM
- cytokines
- actin stress fibers
Apo(a) Stimulates Stress Fiber Formation and VE-Cadherin Dispersion in a Lysine-Dependent Manner

Cho T, et al. JBC 2008
Concentration-Dependent Effects of Apo(a)/Lp(a) on EC Function

- **Vascular protection:**
  - Ox-PL scavenging

- **Wound healing:**
  - ↓ Pericellular Pg activation
  - ↑ Migration
  - ↑ Proliferation
  - ↑ fibrin

- **Atherogenesis:**
  - ↑ Permeability
  - ↑ β-catenin translocation
  - ↑ COX-2
  - ↑ IL-8

- **Atherogenesis:**
  - ↑ Apoptosis
  - ↓ NO
  - ↓ eNOS
  - ↑ ICAM
  - ↑ E-selectin

Physiological role?
Lp(a): Looking Forward

- Lp(a) is entering a new era of clinical relevance

- Can knowledge of Lp(a) production be used to design therapeutics to specifically lower Lp(a)?

- OxPL may hold the key to the pathogenic role of Lp(a) in atherosclerosis
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