Immunomodulatory Pathways in Atherosclerosis

Ziad Mallat, MD, PhD

Department of Medicine, University of Cambridge, Cambridge, UK

Inserm U970
Targeting the Immuno-Inflammatory Response In Atherosclerosis

Moore KJ & Tabas I. Cell 2011
Effect of CC Chemokine Receptor 2 CCR2 Blockade on Serum C-Reactive Protein in Individuals at Atherosclerotic Risk and With a Single Nucleotide Polymorphism of the Monocyte Chemoattractant Protein-1 Promoter Region

Jim Gilbert, MD\textsuperscript{a,*}, Julie Lekstrom-Himes, MD\textsuperscript{b}, Debra Donaldson, MD\textsuperscript{c}, Yih Lee, PhD\textsuperscript{d}, Mingxiu Hu, PhD\textsuperscript{d}, Jing Xu, PhD\textsuperscript{d}, Tim Wyant, MD\textsuperscript{d}, and Michael Davidson, MD\textsuperscript{c,r}, for the MLN1202 Study Group\textsuperscript{t} (Am J Cardiol 2011;107:906–911)

![Graph showing % Change from Baseline CRP over Days]

\( *P < .05 \)
Inhibition of lipoprotein-associated phospholipase A₂ reduces complex coronary atherosclerotic plaque development

Wilensky RL et al., Nat Med 2008
Effects of the Direct Lipoprotein-Associated Phospholipase A₂ Inhibitor Darapladib on Human Coronary Atherosclerotic Plaque

Serruys PW et al., Circulation 2008
Study NCT00799903  Information provided by GlaxoSmithKline

Brief Title
The Stabilization of Atherosclerotic Plaque by Initiation of Darapladib Therapy Trial

Official Title
LPL100601, A Clinical Outcomes Study of Darapladib Versus Placebo in Subjects With Chronic Coronary Heart Disease to Compare the Incidence of Major Adverse Cardiovascular Events (MACE)

Brief Summary
This study will test whether darapladib can safely lower the chances of having a cardiovascular event (such as a heart attack or stroke) in people with coronary heart disease.

Detailed Description
Subjects who qualify for the study will be randomized 1:1 to either darapladib or placebo administered in addition to standard therapy. Following the baseline visit, subjects will be expected to return for clinic visits at 1 month, 3 months, and every 6 months until the end of the study. Average time in the study for an individual subject is expected to be about 3 years.

Study Type
Interventional

Study Phase
Phase III

Current Primary Outcome Measures
Time to the first occurrence of any component of the composite of Major Adverse Cardiovascular Events [MACE: CV death (death due to a cardiovasacular cause), non-fatal myocardial infarction, non-fatal stroke] [ Time Frame: Patients will remain in the study until a specified number of MACE events have occurred. It is anticipated that patients will be in the study about 3 years. ] [ Designated as safety issue: No ]
The Stabilization Of pLaques using Darapladib-Thrombolysis In Myocardial Infarction 52 Trial (SOLID-TIMI 52)

Study NCT01000727  Information provided by GlaxoSmithKline

Brief Title
The Stabilization Of pLaques using Darapladib-Thrombolysis In Myocardial Infarction 52 Trial

Official Title
A Clinical Outcomes Study of Darapladib Versus Placebo in Subjects Following Acute Coronary Syndrome to Compare the Incidence of Major Adverse Cardiovascular Events (MACE).

Brief Summary
This study will test whether darapladib can safely lower the chances of having a cardiovascular event (such as a heart attack or stroke) when treatment is started within 30 days after an acute coronary syndrome (also called ACS).

Detailed Description
Subjects who qualify for the study will be randomized 1:1 to either darapladib or placebo administered in addition to standard therapy. Following the baseline visit, subjects will be expected to return for clinic visits at 1 month, 3 months, 6 months and every 6 months until the end of the study.

Study Type
Interventional

Study Phase
Phase III

Current Primary Outcome Measures
Time to the first occurrence of any component of the composite of Major Adverse Cardiovascular Events [MACE: CV death (death due to a cardiovascular cause), non-fatal myocardial infarction, non-fatal stroke]. [ Time Frame: Through the end of the study. ] [ Designated as safety issue: No ]
Cardiovascular Risk Reduction Study (Reduction in Recurrent Major CV Disease Events)

This study is currently recruiting participants.
Verified November 2011 by Novartis

Study NCT01327846  Information provided by Novartis

Brief Title  ICMJE
Cardiovascular Risk Reduction Study (Reduction in Recurrent Major CV Disease Events)

Official Title  ICMJE
A Randomized, Double-blind, Placebo-controlled, Event Driven Trial of Quarterly Subcutaneous Canakinumab in the Prevention of Recurrent Cardiovascular Events Among Stable Post-myocardial Infarction Patients With Elevated hsCRP

Brief Summary
The purpose of this trial is to test the hypothesis that canakinumab treatment of patients with MI at least one month prior to study entry and elevated hsCRP will prevent recurrent cardiovascular events.

Detailed Description

Study Type  ICMJE
Interventional

Study Phase
Phase III

Study Design  ICMJE
Allocation: Randomized
Intervention Model: Parallel Assignment
Masking: Double Blind (Subject, Investigator, Outcomes Assessor)
Primary Purpose: Treatment

Current Primary Outcome Measures
Time to first occurrence of a major adverse cardiovascular event, which is a composite endpoint consisting of cardiovascular death, non-fatal MI, and stroke. [ Time Frame: 36 Months ] [ Designated as safety issue: No ]

Current Secondary Outcome Measures  ICMJE (submitted: March 31, 2011)
- Time to the first occurrence of the composite cardiovascular endpoint consisting of cardiovascular death, non-fatal MI, stroke, and hospitalization for unstable angina requiring unplanned revascularization. [ Time Frame: 36 Months ] [ Designated as safety issue: No ]
- Time to new onset type 2 diabetes among those with pre-diabetes at randomization. [ Time Frame: 36 Months ] [ Designated as safety issue: No ]
- Time to first occurrence of non-fatal MI, stroke, and all-cause mortality composite. [ Time Frame: 36 Months ] [ Designated as safety issue: No ]
- Time to all-cause mortality. [ Time Frame: 36 Months ] [ Designated as safety issue: No ]
Effects of Interleukin-1β Inhibition With Canakinumab on Hemoglobin A1c, Lipids, C-Reactive Protein, Interleukin-6, and Fibrinogen

A Phase IIb Randomized, Placebo-Controlled Trial

Ridker PM et al. Circulation 2011
Genetic inactivation of IL-1 signaling enhances atherosclerotic plaque instability and reduces outward vessel remodeling in advanced atherosclerosis in mice

Matthew R. Alexander, Christopher W. Moehle, Jason L. Johnson, Zhengyu Yang, Jae K. Lee, Christopher L. Jackson, and Gary K. Owens

rbc (TER-119)  Movat stain  Macrophages (Mac2)  SMCs (SM α-actin)  Collagen (picirsirius red)
Testing the inflammatory hypothesis of atherothrombosis: scientific rationale for the cardiovascular inflammation reduction trial (CIRT)

PAUL M. RIDKER

<table>
<thead>
<tr>
<th>Cohort</th>
<th>Group</th>
<th>HR* (95% CI)</th>
<th>Endpoint</th>
<th>Exposure</th>
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</thead>
<tbody>
<tr>
<td>Wichita Choi 2002</td>
<td>RA</td>
<td>0.4 (0.2–0.8)</td>
<td>Total mortality</td>
<td>LDM</td>
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<tr>
<td></td>
<td></td>
<td>0.3 (0.2–0.7)</td>
<td>CV mortality</td>
<td>LDM</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.4 (0.3–0.8)</td>
<td>CV mortality</td>
<td>LDM &lt; 15 mg wk⁻¹</td>
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<tr>
<td>Netherlands van Helm 2006</td>
<td>RA</td>
<td>0.3 (0.1–0.7)</td>
<td>CVD</td>
<td>LDM only</td>
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<td>0.2 (0.1–0.5)</td>
<td>CVD</td>
<td>LDM + SSZ</td>
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<td></td>
<td>0.2 (0.1–1.2)</td>
<td>CVD</td>
<td>LDM + HCQ</td>
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<tr>
<td></td>
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<td>0.2 (0.1–0.5)</td>
<td>CVD</td>
<td>LDM + SSZ + HCQ</td>
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<td>Miami VA Pradanovich 2005</td>
<td>Psoriasis</td>
<td>0.7 (0.6–0.9)</td>
<td>CVD</td>
<td>LDM</td>
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<tr>
<td></td>
<td>RA</td>
<td>0.5 (0.3–0.8)</td>
<td>CVD</td>
<td>LDM &lt; 15 mg wk⁻¹</td>
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<tr>
<td></td>
<td></td>
<td>0.6 (0.5–0.8)</td>
<td>CVD</td>
<td>LDM</td>
</tr>
<tr>
<td>CORRONA Solomon 2006</td>
<td>RA</td>
<td>0.6 (0.3–1.2)</td>
<td>CVD</td>
<td>LDM TNF-inhibitor</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.4 (0.2–0.8)</td>
<td>CVD</td>
<td>LDM</td>
</tr>
<tr>
<td>QUEST-RA Narango 2008</td>
<td>RA</td>
<td>0.85 (0.8–0.9)</td>
<td>CVD</td>
<td>LDM</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.82 (0.7–0.9)</td>
<td>MI</td>
<td>LDM</td>
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<tr>
<td></td>
<td></td>
<td>0.89 (0.8–1.0)</td>
<td>Stroke</td>
<td>LDM</td>
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<tr>
<td>UK Norfolk 2008</td>
<td>RA, PSA</td>
<td>0.6 (0.4–1.0)</td>
<td>Total mortality</td>
<td>LDM</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.5 (0.3–1.1)</td>
<td>CV mortality</td>
<td>LDM</td>
</tr>
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</table>

to allocate 7000 stable coronary artery disease patients with persistent elevations of hsCRP to placebo or very-low-dose-methotrexate (VLDM, 10 mg weekly), a proven anti-inflammatory regimen that reduces TNFα, IL-6, and CRP levels and is in wide use among rheumatoid arthritis patients.
Peptide-based Therapeutic Vaccination

Auto-Immunity

T_{reg} 

Overwhelm

Antigen-specific memory

Therapeutic Tolerance

? 

Expansion 

T_{reg} 

Peptide therapy

Downregulate

Deletion or anergy

Antigen-specific memory

Naive T cell pool

Larché M & Wraith DC
Nat Med 2005
Inducing tolerance to lipoproteins: Atherosclerosis Vaccine?

ApoB peptide-based vaccination reduces atherosclerosis by inducing a specific Treg cell response.
Regulatory T-Cell Response to Apolipoprotein B100–Derived Peptides Reduces the Development and Progression of Atherosclerosis in Mice

Olivier Herbin, Hafid Ait-Oufella, Wang Yu, Gunilla Nordin Fredriksson, Benjamin Aubier, Nicolas Perez, Véronique Barateau, Jan Nilsson, Alain Tedgui, Ziad Mallat

(Artiosclerosis Thromb Vasc Biol. 2012;32:00-00.)
Immunization of low density lipoprotein (LDL) receptor-deficient rabbits with homologous malondialdehyde-modified LDL reduces atherogenesis

(modified lipoproteins/oxidation/autoantibodies/atherosclerosis/immune system)

Wulf Palinski, Elizabeth Miller, and Joseph L. Witztum

Recombinant Human Antibodies Against Aldehyde-Modified Apolipoprotein B-100 Peptide Sequences Inhibit Atherosclerosis

Alexandru Schiopu, MD; Jenny Bengtsson, PhD; Ingrid Söderberg, BSI; Sabina Janciauskiene, PhD; Stefan Lindgren, MD, PhD; Mikko P.S. Ares, PhD; Prediman K. Shah, MD; Roland Carlsson, PhD; Jan Nilsson, MD, PhD; Gunilla Nordin Fredrikson, PhD

Circulation. 2004
B cell pathways in atherosclerosis

A Study to Evaluate the Safety, Tolerability, and Activity of Intravenous MLDL1278A in Patients on Standard-of-Care Therapy for Stable Atherosclerotic Cardiovascular Disease

**This study is currently recruiting participants.**

Verified November 2011 by Genentech

Study NCT01258907  Information provided by Genentech

<table>
<thead>
<tr>
<th>Brief Title</th>
<th>A Study to Evaluate the Safety, Tolerability, and Activity of Intravenous MLDL1278A in Patients on Standard-of-Care Therapy for Stable Atherosclerotic Cardiovascular Disease</th>
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</thead>
<tbody>
<tr>
<td>Official Title</td>
<td>A Multicenter, Randomized, Double Blind, Placebo-Controlled Phase II Study to Evaluate the Safety, Tolerability, and Activity of Intravenous MLDL1278A in Patients on Standard-of-Care Therapy for Stable Atherosclerotic Cardiovascular Disease</td>
</tr>
<tr>
<td>Brief Summary</td>
<td>This is a Phase II (proof-of-activity), double-blind, placebo-controlled, randomized, multicenter study of MLDL1278A (also known as BI-204) involving patients on standard-of-care therapy for atherosclerotic cardiovascular disease with evidence of vascular inflammation, as quantified by FDG-PET/CT.</td>
</tr>
<tr>
<td>Study Type</td>
<td>Interventional</td>
</tr>
<tr>
<td>Study Phase</td>
<td>Phase II</td>
</tr>
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</table>
| Study Design | Allocation: Randomized  
Intervention Model: Parallel Assignment  
Masking: Double Blind (Subject, Investigator)  
Primary Purpose: Treatment |
| Current Primary Outcome Measures | Change in TBR as measured by FDG-PET/CT [ Time Frame: Baseline to Week 12 ]  
[ Designated as safety issue: No ] |
| Current Secondary Outcome Measures | Incidence and severity of adverse events and clinical laboratory abnormalities as a measure of safety and tolerability of MLDL1278A [ Time Frame: Throughout study or until early discontinuation ]  
[ Designated as safety issue: No ] |

(Submitted: December 10, 2010)
They respond to T cell–dependent antigens and contribute to adaptive immunity

They respond to T cell–indendependent antigens, selectivity for self antigens, producers of natural IgM

Mackay F
*Nat Rev Immunol* 2009
CD20 mAb-mediated B Cell Depletion in Atherosclerosis
Decreased T cell infiltration after anti-CD20 therapy
B cell depletion reduces the development of atherosclerosis in mice

Ait-Oufella et al.

JEM 2010
BAFF – B cell activating factor

**B Cell Physiology**
- B2 cell survival
- Class switching
- Plasma Cells
- Autoreactive B cells

**Autoimmune Diseases**
- SLE
- Rheumatoid Arthritis
- Sjogren’s Syndrome
- Anti-Phospholipid syndrome

**T Cells**
- Myeloid cells

**Atherosclerosis?**
BAFF Levels in Human Carotid Lesions and Relation with Plaque Phenotype

P<0.001
BAFF Receptor Deficiency Reduces the Development of Atherosclerosis in Mice

A

% Lymphocytes

WT

BAFF-R⁻/⁻

BM

Sp

BI

LN

B

% Lymphocytes

B1a

CD5⁺CD11b⁺

B1b

B2

C

RLU (x10⁴)

IgM

IgG₁

IgG₂c

D

% CD68⁺CD4⁺ T Cells

% CD44hi CD4⁺ T Cells

E

CPM (x10³)

CD4⁺ T eff

*"
Lesion Size

B cell-selective BAFF-R deletion

Macrophages

T lymphocytes
Acute Myocardial Infarction

Evolution of therapies in the management of acute coronary syndromes


Aspirin, Low molecular weight heparin, Heparin, Ilb/IIa receptor antagonist, Clopidogrel, Atorvastatin, Fondaparinux, Bivalirudin, Integrated strategy

Post-MI Inflammation

Blood

Ischemic Myocardium

Chemokines release (CCL2, MIP-1α, CX3CL1)

LCA occlusion

Neutrophils

CD11b+

Ly6G+

Inflammatory Monocytes

Ly6C<sup>hi</sup>

CCR2+

7/4<sup>hi</sup>

CX3CR1<sup>lo</sup>

Resident Monocytes

Ly6C<sup>lo</sup>

CCR2-

7/4<sup>lo</sup>

CX3CR1<sup>hi</sup>

Pro-Inflammatory cytokines

High Proteolysis

Anti-inflammatory cytokines

Angiogenesis

Apoptosis

Adverse remodelling

Myocardial healing
Role of B lymphocytes in immuno-inflammatory response and tissue remodelling after myocardial infarction
Kinetics of inflammatory cell infiltration after Myocardial Infarction

Digestion (Col I, XI, Hyaluronidase, DNase) → Gradient Density Centrifugation → FACS Analysis

**Neutrophils**
*(CD11b+ Ly6G+ 7/4^{HI})*

- MI
- sham

**7/4^{HI} and 7/4^{LOW} Monocytes**

- 7/4^{HI} MI
- 7/4^{HI} MI sham
- 7/4^{LOW} MI
- 7/4^{LOW} MI sham

**CD3+ T lymphocytes**

- MI
- sham
Kinetics of B lymphocytes infiltration after Myocardial Infarction

- **Flow cytometry analysis**

  ![Flow cytometry analysis](image1)

- **B220 Immunostaining**

  ![B220 Immunostaining](image2)
Anti-CD20 antibody treatment reduces B lymphocyte levels

- Blood
  - Mature B cells
  - 200 μg/mouse 1 hour after MI

- Spleen D14
  - B220<sup>Hi</sup> IgM<sup>+</sup>

- Heart D3
  - B220<sup>Hi</sup> IgM<sup>+</sup>

- Heart D5
  - B220<sup>Hi</sup> IgM<sup>+</sup>
B lymphocyte depletion limits adverse LV remodelling

- **LV internal dimension in diastole**
  - PBS
  - Anti-CD20
  - D14

- **LV Posterior Wall Thickness in systole**
  - PBS
  - Anti-CD20
  - D14

- **Shortening Fraction**
  - PBS
  - Anti-CD20
  - D14

- **Infarct Size (Masson Trichrome staining)**
  - PBS
  - Anti-CD20
  - D14
B lymphocyte depletion reduces systemic and local post-MI inflammation

- **Spleen D14**

- **Heart D14**
B lymphocyte depletion alters monocyte distribution in post-MI setting

**Bone Marrow D3**

- **PBS D3**
  - CD11b vs. 7/4 (Ly6-C)
  - Monocytes $7/4^{\text{HI}}$
  - Monocytes $7/4^{\text{Lo}}$

- **α-CD20 D3**
  - CD11b vs. 7/4 (Ly6-C)
  - Monocytes $7/4^{\text{HI}}$
  - Monocytes $7/4^{\text{Lo}}$

**Blood D3**

- **PBS D3**
  - CD11b vs. 7/4 (Ly6-C)
  - Monocytes $7/4^{\text{HI}}$
  - Monocytes $7/4^{\text{Lo}}$

- **α-CD20 D3**
  - CD11b vs. 7/4 (Ly6-C)
  - Monocytes $7/4^{\text{HI}}$
  - Monocytes $7/4^{\text{Lo}}$
B lymphocyte depletion selectively reduces MCP-3 levels in post-MI

- **MCP-1/CCL2**
  - Not Detected in B cell supernatants

- **MCP-3/CCL7**
  - MCP-1/CCL2
  - Not Detected in B cell supernatants
B lymphocytes trigger 7/4^{Hi} monocytes migration

**7/4^{Hi} Monocytes transmigration assay**

**Lower compartment:**
- Medium RPMI 10% SVF
- B cells (2.10^6)
- α-CD40 and IgM-treated B lymphocytes

![Image of transwell insert with 7/4^{Hi} monocytes and B lymphocytes](image)

Control
B cells
Activated B cells
αMCP1
αMCP3

Control
B cells
Activated B cells
Activated B cells +αMCP1
Activated B cells +αMCP3

7/4^{Hi} Monocytes migration (% of control)

***

***

***
Exogenous administration of B lymphocytes promotes adverse LV remodelling

Injection splenocytes WT, B cell-depleted splenocytes ± WT or MCP-3-deficient B cells

Rag1⁻/⁻

B cells levels

B cells/spleen [x10⁶]

Spleen WT

Spleno αCD20

Spleno αCD20 + WT B cells

Spleno αCD20 + MCP3⁻/⁻ B cells

D-7 D0 D3 D14

MI FACS Echocardiography
Exogenous administration of B lymphocytes enhances $7/4^{Hi}$ monocytes mobilisation and infiltration into the ischemic heart.
Exogenous administration of B lymphocytes promotes adverse LV remodelling.
FAST-MI is a nationwide French registry carried out in 3059 consecutive pts with AMI admitted in 223 CCUs.

100 centers, which included 1036 patients, participated in the serum databank.

Outcome events were defined as all-cause death, recurrent AMI and incident stroke.

The 24-month follow-up of mortality was complete for 95% of patients. 170 events occurred during follow-up.
B Lymphocytes Trigger CCL7-Dependent Monocyte Mobilisation and Promote Adverse Ventricular Remodelling after Acute Myocardial Infarction

B cell depleting and CCL7-targeting therapies may be cardioprotective

Zouggari Y et al., *Nature Medicine*, In Press
Treating inflammation in atherosclerotic cardiovascular disease: emerging therapies

Roland Klingenberg\textsuperscript{1,2*} and Göran K. Hansson\textsuperscript{2}

European Heart Journal (2009) 30, 2838–2844

**Pathology**
- Healthy artery
- Fatty streak
- Atherosclerotic plaque
- Plaque rupture or erosion + thrombus formation

**Clinical stage**
- Asymptomatic
- Asymptomatic or angina (CCS I-IV)
- ACS

**Anti-inflammatory therapies:**
- **Current:** Statins
- **Experimental:** Immunization

**PLA2 inhibitors**
- Anti-IL1beta

**Peptide-based vaccination**
- B2 cell depleting agents
University of Cambridge
British Heart Foundation

Andy SAGE
Xuan Li
Deirdre MURPHY
Lauren BAKER
James HARRISON
Leanne MASTERS

Inserm U970
Alain TEDGUI
Hafid AIT-OUFELLA
Olivier HERBIN
Patrick BRUNEVAL

Jean-Sébastien SILVESTRE
Yasmine ZOUGGARI

Duke University, USA
Thomas F. TEDDER

UCSF, USA
Israel F. CHARO

CeMM, Medical University of Vienna, Austria
Christoph J. BINDER

AP-HP, Pierre et Marie Curie University
Tabassome SIMON

University of Utrecht, The Netherlands
Gerard PASTERKAMP
PLA2 enzymes hydrolyze phospholipids at the *sn-2* position to generate lysophospholipids and fatty acids.
Lipoprotein-Associated and Secreted Phospholipases A₂ in Cardiovascular Disease
Roles as Biological Effectors and Biomarkers

Ziad Mallat, MD, PhD; Gérard Lambeau, PhD; Alain Tedgui, PhD
Lipoprotein-Associated Phospholipase A\textsubscript{2} Protein Expression in the Natural Progression of Human Coronary Atherosclerosis

Kolodgie et al., Arterioscler Thromb Vasc Biol 2006
Lipoprotein-associated phospholipase A₂ and risk of coronary disease, stroke, and mortality: collaborative analysis of 32 prospective studies

The Lp-PLA₂ Studies Collaboration

Lancet 2010; 375: 1536-44

RR (95% CI) per 1 SD higher

<table>
<thead>
<tr>
<th>Parameter</th>
<th>RR (95% CI)</th>
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<tbody>
<tr>
<td>Lp-PLA₂ activity</td>
<td>1.11 (1.06–1.16)</td>
</tr>
<tr>
<td>Lp-PLA₂ mass</td>
<td>1.11 (1.07–1.15)</td>
</tr>
<tr>
<td>Systolic blood pressure</td>
<td>1.10 (1.00–1.22)</td>
</tr>
<tr>
<td>Smoking status*</td>
<td>1.34 (1.19–1.51)</td>
</tr>
<tr>
<td>Non-HDL cholesterol</td>
<td>1.10 (1.02–1.18)</td>
</tr>
<tr>
<td>HDL cholesterol†</td>
<td>1.15 (1.05–1.25)</td>
</tr>
</tbody>
</table>
Oxidation-Specific Biomarkers, Lipoprotein(a), and Risk of Fatal and Nonfatal Coronary Events

Sotirios Tsimikas, MD,* Ziad Mallat, MD, PhD,† Philippa J. Talmud, DSC,‡ John J. P. Kastelein, MD, PhD,¶ Nicholas J. Wareham, MBBS, PhD,§ Manjinder S. Sandhu, PhD,‖ Elizabeth R. Miller, BS,* Joelle Benessiano, MD, PhD,† Alain Tedgui, PhD,‡ Joseph L. Witztum, MD,* Kay-Tee Khaw, MBBS, PhD,‖ S. Matthijs Boekholdt, MD, PhD¶

La Jolla, California; Paris, France; London and Cambridge, United Kingdom; and Amsterdam, the Netherlands

Results

After adjusting for age, smoking, diabetes, low- and high-density lipoprotein cholesterol, and systolic blood pressure, the highest tertiles of oxidized phospholipids on apolipoprotein B-100 particles and Lp(a) were associated with a significantly higher risk of CAD events (odds ratios: 1.67 and 1.64, respectively; p < 0.001) compared with the lowest tertiles. The odds ratio of CAD events associated with the highest tertiles of oxidized phospholipids on apolipoprotein B-100 particles or Lp(a) was significantly potentiated (approximately doubled) by the highest tertiles of secretory phospholipase A2 activity and mass but less so for myeloperoxidase and lipoprotein-associated phospholipase A2 activity. The odds ratios for fatal CAD were higher than for the combined end point. After taking into account the Framingham Risk Score, c-index values progressively increased when oxidative biomarkers were added to the model.

Conclusions

This EPIC-Norfolk study links pathophysiologically related oxidation-specific biomarkers and Lp(a) with CAD events. Oxidation-specific biomarkers provide cumulative predictive value when added to traditional cardiovascular risk factors. (J Am Coll Cardiol 2010;56:946–55) © 2010 by the American College of Cardiology
Prognostic Utility of Secretory Phospholipase A₂ in Patients with Stable Coronary Artery Disease

Michelle L. O’Donoghue,¹* Ziad Mallat,²,³ David A. Morrow,¹ Joelle Benessiano,⁴ Sarah Sloan,¹ Torbjørn Omland,⁵ Scott D. Solomon,⁶ Eugene Braunwald,¹ Alain Tedgui,²,³ and Marc S. Sabatine¹

BACKGROUND: Secretory phospholipase A₂ (sPLA₂) may contribute to atherogenesis. To date, few prospective studies have examined the utility of sPLA₂ for risk stratification in coronary artery disease (CAD).

RESULTS: After multivariable adjustment, sPLA₂ was associated with an increased risk of cardiovascular death, myocardial infarction, or stroke (adjusted hazard ratio Q4:Q1 1.55, 95% CI 1.13–2.14) and cardiovascular death or heart failure (1.91, 1.20–3.03). In further multivariable assessment, increased activity levels of sPLA₂ were associated with the risk of cardiovascular death, myocardial infarction, or stroke (adjusted hazard ratio 1.47, 95% CI 1.06–2.04), independent of lipoprotein-associated phospholipase A₂ mass and C-reactive protein, and modestly improved the area under the curve (AUC) beyond established clinical risk factors (AUC 0.668–0.675, P = 0.01). sPLA₂, N-terminal pro-B-type natriuretic peptide, and high-sensitivity cardiac troponin T all were independently associated with cardiovascular death or heart failure, and each improved risk discrimination (P = 0.02, P < 0.001, P < 0.001, respectively).

METHODS: We measured plasma sPLA₂ activity at baseline in 3708 subjects in the PEACE randomized trial of trandolapril vs placebo in stable CAD. Median follow-up was 4.8 years. We used Cox regression to adjust for demographics, clinical risk factors, apolipoprotein B, apolipoprotein A1, and medications.

CONCLUSIONS: sPLA₂ activity provides independent prognostic information beyond established risk markers in patients with stable CAD. These data are encouraging for studies designed to evaluate the role of sPLA₂ as a therapeutic target.

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Type II Secretory Phospholipase A₂ and Prognosis in Patients with Stable Coronary Heart Disease: Mendelian Randomization Study

Lutz P. Breitling¹*, Wolfgang Koenig², Marcus Fischer³, Ziad Mallat⁴, Christian Hengstenberg³, Dietrich Rothenbacher¹,⁵, Hermann Brenner¹

Abstract

Background: Serum type II secretory phospholipase A₂ (sPLA₂-IIa) has been found to be predictive of adverse outcomes in patients with stable coronary heart disease. Compounds targeting sPLA₂-IIa are already under development. This study investigated if an association of sPLA₂-IIa with secondary cardiovascular disease (CVD) events may be of causal nature or mainly a matter of confounding by correlated cardiovascular risk markers.

Methodology/Principal Findings: Eight-year follow-up data of a prospective cohort study (KAROLA) of patients who underwent in-patient rehabilitation after an acute cardiovascular event were analysed. Associations of polymorphisms (SNP) in the sPLA₂-IIa-coding gene PLA₂G2A with serum sPLA₂-IIa and secondary fatal or non-fatal CVD events were examined by multiple regression. Hazard ratios (HR) were compared with those expected if the association between sPLA₂-IIa and CVD were causal. The strongest determinants of sPLA₂-IIa (rs4744 and rs10732279) were associated with an increase of serum concentrations by 81% and 73% per variant allele. HRs (95% confidence intervals) estimating the associations of the SNPs with secondary CVD events were increased, but not statistically significant (1.16 [0.89–1.51] and 1.18 [0.91–1.52] per variant allele, respectively). However, these estimates were very similar to those expected when assuming causality (1.18 and 1.17), based on an association of natural log-transformed sPLA₂-IIa concentration with secondary events with HR = 1.33 per unit.

Conclusion: The present findings regarding genetic polymorphisms, determination of serum sPLA₂-IIa, and prognosis in CVD patients are consistent with a genuine causal relationship and thus might point to a valid drug target for prevention of secondary CVD events.
sPLA2 inhibitor acts synergistically with statin to decrease atherosclerosis

apoE-/- mice treated with varespladib (A-002) +/- pravastatin

Effects of 1-H-indole-3-glyoxamide (A-002, Varespladib, Anthera Pharmaceuticals) on concentration of sPLA2 (PLASMA study): a phase II double-blind, randomised, placebo-controlled trial

393 patients randomly assigned received placebo (n=79) or the sPLA2 inhibitor, A-002: 50 mg (n=79), 100 mg (n=80), 250 mg (n=78), or 500 mg (n=77) twice daily, for 8 weeks.

Primary endpoint:
change in sPLA2 IIA concentration or activity from baseline to week 8

Results: Dose dependent reduction in sPLA2-IIA concentration in the A-002 groups (from $69 \pm 2\%$ in the 50 mg group to $95 \pm 8\%$ in the 500 mg group), significantly different from placebo ($p<0.0001$)
VISTA-16 Trial: Evaluation of Safety and Efficacy of Short-term A-002 Treatment in Subjects With Acute Coronary Syndrome

This study is currently recruiting participants.
Verified January 2012 by Anthera Pharmaceuticals

Study NCT01130246  Information provided by Anthera Pharmaceuticals

Detailed Description

A double-blind randomized parallel group placebo controlled study in subjects presenting with an ACS. Up to 6500 subjects will be randomized to receive either A-002 500 mg once daily (QD) or placebo tablets in addition to atorvastatin QD and standard of care. Treatment will be 16 weeks in duration. The dose of atorvastatin shall be adjusted after 8 weeks if subject's LDL-C is \( \geq 100 \) mg/dL, but otherwise must remain stable throughout the 16-week duration of study. The survival status for all enrolled subjects will be ascertained 6 months after they complete the study.

Randomization must occur within \( \leq 96 \) hours of hospitalization for the index ACS event, or if already hospitalized, within \( \leq 96 \) hours of index event diagnosis. Follow-up visits will occur on Weeks 1, 2, 4, 8, and 16. A 6 month follow-up visit will also occur.

The primary objective of the study is to determine whether 16 weeks of treatment with A-002 plus atorvastatin and standard of care is superior to placebo plus atorvastatin and standard of care for reducing the hazard of the first occurrence of the combined endpoint of cardiovascular death, non-fatal myocardial infarction, non-fatal stroke, or documented unstable angina with objective evidence of ischemia requiring hospitalization.
<table>
<thead>
<tr>
<th>Genotype/haplotype</th>
<th>Population (size)</th>
<th>Endpoint</th>
<th>p value</th>
<th>RR/OR</th>
<th>Reference</th>
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<tbody>
<tr>
<td>Add/del allele 5-LO promoter (ALOX5)</td>
<td>USA (Los Angeles; 470)</td>
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<tr>
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<td>Stroke</td>
<td>NS</td>
<td>–</td>
<td>94</td>
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<tr>
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<tr>
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<tr>
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<tr>
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<td></td>
<td>0.024</td>
<td>1.26</td>
<td>94</td>
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</tbody>
</table>
Treatment With 5-Lipoxygenase Inhibitor VIA-2291 (Atreleuton) in Patients With Recent Acute Coronary Syndrome

Jean-Claude Tardif, MD; Philippe L. L’Allier, MD; Reda Ibrahim, MD; Jean C. Grégoire, MD; Anna Nozza, MSc; Mariève Cossette, MSc; Simon Kouz, MD; Marc-André Lavoie, MD; Janie Paquin, RT; Tilmann M. Brotz, PhD; Rebecca Taub, MD; Josephine Pressacco, MD, PhD
B lymphocyte depletion limits adverse LV remodelling

- **Apoptotic cells number (Tunel)**
  - PBS
  - Anti-CD20

- **Capillary Density (BS1 lectin, WGA)**
  - PBS
  - Anti-CD20

- **Arteriolar Density (α-actin)**
  - PBS
  - Anti-CD20
MCP-3 deficiency preserves LV function after acute MI

**Monocytes 7/4**

- **Blood D0**
  - WT
  - MCP3KO

- **Blood D14**
  - WT
  - MCP3KO

- **BM D14**
  - WT
  - MCP3KO

- **Shortening Fraction**

- **D14**
  - PBS
  - Anti-CD20

* *