1. What lipoproteins are represented by unique, but related, lipid and lipoprotein biomarkers?

2. What is the impact of discordance between alternate lipid and lipoprotein measures?

3. What evidence is needed to support utilization of alternate lipid and lipoprotein measures for risk assessment versus risk management?
**Relationship of Cholesterol and ApoB Lipoproteins**

<table>
<thead>
<tr>
<th>Total Cholesterol</th>
<th>All Apo B-Containing Lipoproteins</th>
</tr>
</thead>
<tbody>
<tr>
<td>GOOD HDL</td>
<td>BAD</td>
</tr>
<tr>
<td>Apo A</td>
<td>Apo B</td>
</tr>
<tr>
<td>LDL-C</td>
<td>Non-HDL-C</td>
</tr>
</tbody>
</table>

**Why are non-HDL-C and ApoB more strongly associated with CHD risk than LDL-C?**

A. Benefit from inclusion of non-LDL ApoB containing lipoproteins

B. Better represent LDL quantity than LDL-C

**Cholesterol / Ester Triglyceride**

**Apo A**

**Apo B**

**Relationship of Alternate Measures of Atherogenic Lipoproteins with Future CVD Events**

Framingham (431 events)

<table>
<thead>
<tr>
<th>Cholesterol Measures</th>
<th>HR (95% CI)</th>
<th>p</th>
</tr>
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<tr>
<td>LDL-C</td>
<td>1.11 (1.01-1.22)</td>
<td>0.03</td>
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<tr>
<td>Non-HDL-C</td>
<td>1.21 (1.10-1.33)</td>
<td>&lt;0.0001</td>
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Values are from multivariable logistic regression analyses adjusted for age, gender, BP, smoking, and lipid vs. Hazard ratio (HRs) are per 1 SD increment of the lipoprotein variable.

**Relations of Alternate Measures of Atherogenic Lipoproteins with Future CVD Events**

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**Explanations:**


**Except for type III hyperlipidemia, more than 90% of total plasma ApoB particles are LDL particles (even among high TG patients) [1,2].**

**Evolving Views of LDL**

<table>
<thead>
<tr>
<th>Conventional Explanation</th>
<th>Different Measures of Atherogenic Lipoproteins</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>LDL</td>
</tr>
<tr>
<td></td>
<td>LDL-C</td>
</tr>
<tr>
<td></td>
<td>apoB</td>
</tr>
</tbody>
</table>

**Contemporary View**

<table>
<thead>
<tr>
<th>4 Different Ways to Assess LDL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cholesterol Content</td>
</tr>
<tr>
<td>---------------------</td>
</tr>
<tr>
<td>LDL-C</td>
</tr>
<tr>
<td>Non-HDL-C</td>
</tr>
</tbody>
</table>

**Evaluation of Individual Biomarker Outcome Associations**

- **Total Population**

- **Hazard Ratio**
  - **Highest Quintile**
  - **Lowest Quintile**
Impact of Discordance Between Alternate Measures on Outcome Associations

Total Population
Concordant
Discordant

Where Concordance Is Dominant There Is No Ability To Detect A Difference In Performance Of The New Measure

Concordant No Difference In Outcomes Between Biomarkers

Discordant Potential Significance Difference in Outcomes Between Biomarkers


Discordance Between Alternate LDL Measures
Quebec Cardiovascular Study (n=2,103)

Adapted from Sniderman AD, et al. Am J Cardiol 2003;91:1173-1177
The contradictory results of observational studies and clinical trials in which discordant and concordant subjects have not been separated has led to persistent uncertainty as to whether there is advantage to alternate measures of atherogenic lipoprotein-related risk of vascular disease.

Impact of Discordance Between Alternate Measures on Outcome Associations

CHD Event Associations of LDL-P versus LDL-C
Framingham Offspring Study (n=3,066)

Clinical Implications of Discordance Between Low-Density Lipoprotein Cholesterol and Particle Number

James D. Otvos, PhD, Samia Mora, MD, MHS, Irina Shalaurova, MD, Philip Greenland, MD, Rachel H. Mackey, PhD, MPH, David C. Goff Jr., MD, PhD

Journal of Clinical Lipidology 2011;5:105-113
Study Design
Multi-Ethnic Study of Atherosclerosis (MESA)
• Large NHLBI observational study of the pathogenesis and progression of subclinical atherosclerosis.
• Baseline Lipid and NMR measurements of entire cohort.

Objective
• To compare incident CHD events in subjects with concordant versus discordant LDL-C and LDL-P measures

Outcome Measures
• 319 incident CVD events (incident CVD included myocardial infarction, coronary heart disease death, angina, stroke, stroke death, or other atherosclerotic or CVD death) during 5.5-yr follow-up.

Multi-Ethnic Study of Atherosclerosis (MESA) [6,697]

Analytic Relations of Lipid and Particle Number Measures
Multi-Ethnic Study of Atherosclerosis (MESA) [n=6,697]

From Cox regression analyses adjusted for age, sex, and race.

Multi-Ethnic Study of Atherosclerosis (MESA)
Relations with CVD Events in Concordant/Discordant LDL Subgroups

<table>
<thead>
<tr>
<th>LDL-C</th>
<th>LDL-P</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR</td>
<td>p</td>
</tr>
<tr>
<td>HR</td>
<td>p</td>
</tr>
</tbody>
</table>

Overall: 1.20 0.0009 1.32 <0.0001

CVD Event Rates in Subgroups with Low LDL-C


CVD Event Rates in Subgroups with Low LDL-P


LDL-P and LDL-C Discordance in MESA
Relations with Incident CVD Events (n=319)

Discordance analysis of Apolipoprotein B and non-high density lipoprotein cholesterol as markers of cardiovascular risk in the INTERHEART study

Allan D. Sniderman, Shofique Islam, Salim Yusuf, Matthew J. McQueen

Atherosclerosis 2012;225:444-449

Study Design

Subjects
- Subjects were recruited from 262 centers in 52 countries.
- Blood samples were obtained from 9,345 cases (first acute myocardial infarction) and 12,120 age and sex-matched controls without known cardiovascular disease.

Objective
- To compare the odds ratio of cases to controls for discordant groups compared to the ratio of cases to controls in the concordant group (reference group).
- Values of apoB and non-HDL-C were expressed as percentiles of the study population. Difference > 5 percentile points defined discordance.
- 10,949 (51%) subjects were concordant and 10,516 (49%) were discordant

Relationship of ApoB and Non-HDL Cholesterol

Interheart (n=21,465)

A

B

Odds Ratio 1.48

Odds Ratio 0.72
Conclusions

Interheart (n=21,465)

1. “In summary, LDL-C, non-HDL-C and apoB are closely related metabolic markers of cardiovascular risk. Nevertheless, based on differences in cholesterol content, subjects who are discordant for non-HDL-C and apoB can be distinguished from those who are concordant.”

2. “In those who are concordant, apoB and non-HDL-C will be equivalent markers of risk.”

3. “Our data demonstrate that in those who are discordant, apoB is superior to non-HDL-C.”


Major Lipids, Apolipoprotein, and Risk of Vascular Disease

The Emerging Risk Factors Collaboration Writing Group


Emerging Risk Factors Collaboration

Study Design

- Evaluated the odds ratio (first versus fifth quintiles) for lipid and lipoprotein biomarkers with CHD events (i.e., first-ever MI or fatal CHD).
- Analyses were based on 91,307 participants (involving 4,499 cases) from 22 studies.
- Regression analyses were stratified, where appropriate, by sex and trial group and adjusted for age, systolic blood pressure, smoking status, history of diabetes mellitus, and body mass index.

A Meta-Analysis of Low-Density Lipoprotein Cholesterol, Non-High-Density Lipoprotein Cholesterol, and Apolipoprotein B as Markers of Cardiovascular Risk

Allan D. Sniderman, MD; Ken Williams, MSc; John H. Contois, PhD; Howard M. Monroe, PhD; Matthew J. McQueen, MBChB, PhD; Jacqueline de Graaf, MD, PhD; Curt D. Furberg, MD, PhD


Meta-Analysis of LDL-C, Non-HDL-C, and ApoB as Markers of Cardiovascular Risk

**Study Design:**
- Meta-analysis of all published epidemiologic studies with estimates of relative risks of fatal or nonfatal ischemic cardiovascular events and measures of non-HDL-C and apoB.
- 12 independent reports, including 233,455 subjects and 22,950 events, were analyzed.

**Major Findings:**
- Whether analyzed individually or in head-to-head comparisons, apoB was the most potent marker of cardiovascular risk.

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>ERR</th>
<th>95% Confidence Interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>ApoB</td>
<td>1.43</td>
<td>1.35 – 1.51</td>
</tr>
<tr>
<td>Non-HDL-C</td>
<td>1.34</td>
<td>1.24 – 1.44</td>
</tr>
<tr>
<td>LDL-C</td>
<td>1.25</td>
<td>1.18 – 1.33</td>
</tr>
</tbody>
</table>

**Meta-Analysis of LDL-C, Non-HDL-C, and ApoB as Markers of Cardiovascular Risk**

- “The present analysis indicates that non-HDL-C is superior to LDL-C as a marker of cardiovascular risk.”
- “The conventional explanation would be that the gain in predictive power is due to the cholesterol in VLDL.”
- “The superiority of non-HDL-C over LDL-C is due to the fact that non-HDL-C is a better marker of LDL-P than LDL-C.”
- “When apoB and non-HDL-C are concordant, they will predict risk equally, whereas when they are discordant, apoB will be superior.”


**Association of LDL Cholesterol, Non–HDL Cholesterol, and Apolipoprotein B Levels With Risk of Cardiovascular Events Among Patients Treated With Statins: A Meta-analysis**


**Meta-Analysis of Outcome Association with Alternate LDL Measures on Statin Therapy**

- Individual level patient data obtained from 8 statin trials in which LDL-C, non-HDL-C and ApoB levels were measured at 1 year
- Among 38,153 patients allocated to statin therapy:
  - 156 fatal myocardial infarctions,
  - 1,678 nonfatal myocardial infarctions
  - 615 fatal events from other coronary artery disease
  - 2,806 hospitalizations for unstable angina,
  - 1,029 fatal or nonfatal strokes occurred during follow-up.
- Hazard ratios (HRs) and corresponding 95% CIs for risk of major cardiovascular events adjusted for established risk factors by 1-SD increase in LDL-C, non-HDL-C, and apoB.
Meta-analysis of comparison of effectiveness of lowering apolipoprotein B versus low-density lipoprotein cholesterol and non-high-density lipoprotein cholesterol for cardiovascular risk reduction in randomized trials.

J. G. Robinson, S. Wang and T. A. Jacobson

American Journal of Cardiology 2012;110(10):1468-76.

Meta-Analysis of Outcome Association with Alternate LDL Measures on Statin Therapy

- Study evaluated the relation between apolipoprotein B (apoB) decrease and coronary heart disease, stroke, and cardiovascular disease risk.
- Bayesian random-effects meta-analysis was used to evaluate the association of mean absolute apoB decrease (mg/dL) with relative risk of:
  - CHD (nonfatal myocardial infarction and coronary heart disease death)
  - stroke (nonfatal stroke and fatal stroke)
  - CVD (coronary heart disease, stroke, and coronary revascularization).
Meta-Analysis of Outcome Association with Alternate LDL Measures on Lipid Altering Therapy

- Analysis included 25 trials (n=131,134):
  - 12 statin therapy
  - 4 fibrate therapy
  - 5 niacin therapy
  - 2 simvastatin – ezetimibe therapy
  - 1 on ileal bypass surgery
  - 1 on aggressive versus standard low-density lipoprotein (LDL) cholesterol and blood pressure targets.


Meta-Analysis of Outcome Association with Alternate LDL Measures on Lipid Altering Therapy

Combining the 25 trials:
- Each 10-mg/dl decrease in apoB was associated with a 9% decrease in CHD, no decrease in stroke, and a 6% decrease in major CVD risk.
- non-HDL-C decrease modestly outperformed apoB decrease for prediction of CHD and CVD risk reduction;

In the 12 statin trials:
- apoB and non-HDL-C decreases similarly predicted cardiovascular disease risk;
- apoB decreases added information for predicting CHD beyond LDL-C and non-HDL-C decreases, but did not improve CVA or CVD disease risk prediction.


Evaluating Cardiovascular Biomarkers

<table>
<thead>
<tr>
<th>Intended Application</th>
<th>Type of Biomarker</th>
<th>Clinical Use</th>
<th>Impact on Clinical Decision Making</th>
<th>Evidence Needed to Support Use</th>
</tr>
</thead>
<tbody>
<tr>
<td>Risk Assessment</td>
<td>New Biomarker</td>
<td>Biomarker is additional to information from established risk assessment.</td>
<td>Significant improvement in risk stratification with the addition of new biomarker, or substitution of new measure of existing biomarker, in risk models (net reclassification index, ROC).</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(Inflammatory measures, particle size)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>New Measure of Established Biomarker (LDL Quantity)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Risk Management</td>
<td>New Biomarker</td>
<td>Biomarker serves as a treatment target and guides need for additional therapy</td>
<td>Outcome improvement with new measure in the setting of discordance.</td>
<td></td>
</tr>
<tr>
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<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Recommendations for Using LDL Particle Number Measures as Targets of Therapy

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Population</th>
<th>&lt;5th</th>
<th>20th</th>
<th>50th</th>
<th>80th</th>
</tr>
</thead>
<tbody>
<tr>
<td>LDL-C (mg/dL)</td>
<td>Framingham (1)</td>
<td>&gt; 75</td>
<td>100</td>
<td>120</td>
<td>125</td>
</tr>
<tr>
<td>ApoB (mg/dL)</td>
<td>&gt; 60</td>
<td>80</td>
<td>100</td>
<td>120</td>
<td></td>
</tr>
</tbody>
</table>

Organization | Proposed ApoB Targets of Therapy (mg/dL) |
-------------|-----------------------------------------|
Canadian Cardiovascular Society Guidelines (2) | NA | <80 |
American Diabetes Association / American College of Cardiology Foundation Consensus Statement (3) | <80 | <80 | NA |
American Association for Clinical Chemistry Lipoprotein & Vascular Disease Working Group Recommendations (1) | <80 | <100 | |
American Association of Clinical Endocrinologists Guidelines for Management of Dyslipidemias (4) | <80 | <90 | NA |
European Society of Cardiology (ESC)/European Atherosclerosis Society (EAS) Guidelines for the Management of Dyslipidaemias (5) | <80 | <100 | NA |
National Lipid Association Expert Recommendations (6) | Option: <70 | <80 | <100 |

* NMR LDL-P targets also recommended at population equivalent levels

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**Summary**

1. Alternate cholesterol (LDL-C, non-HDL-C) and lipoprotein particle number (apoB, NMR LDL-P) LDL measures are available for clinical use.
2. Although highly correlated, lipid and particle number measures are frequently discordant indicating that one measure can not easily substitute for another.
3. Contradictory results of observational studies and clinical trials in which discordant and concordant subjects have not been separated has led to persistent uncertainty as to whether there is advantage to alternate measures of atherogenic lipoprotein-related risk of vascular disease.
4. The criteria for judging the clinical utility of alternate measures of an established biomarker is the strength of outcome associations for the new measure in the discordant setting.
5. When cholesterol (LDL-C, non-HDL-C) and particle number (apoB, NMR LDL-P) measures are concordant, each predict outcomes equally. When discordant, apoB and NMR LDL-P are more significantly predictive of CHD outcomes than LDL-C or non-HDL-C.