Causes of death, 2001:

1. Infectious and parasitic diseases: 14.9 million
2. Heart diseases: 11.1 million
3. Cancers: 7.3 million
4. Stroke: 5.5 million
5. Respiratory diseases: 3.6 million
6. Accidents, fires, drowning, etc.: 3.5 million
7. Maternal and perinatal: 3.0 million
8. Violence (war, homicide, suicide): 1.6 million

USA

Population: 6,122,210,000
Deaths: 56,554,000

World Health Organization
World Health Report 2002
Atherosclerosis

Risk increases with higher [LDL]
Risk decreases with higher [HDL]
Familial Hypercholesterolemia (FH)

Heterozygotes (1:500)
- 300-500 mg/dl plasma cholesterol
- Xanthomas in third decade
- Coronary heart disease in fourth decade
- Treat w/ statins and bile acid binding resins

Homozygotes (1:10^6)
- 500-1200 mg/dl plasma cholesterol
- Xanthomas at birth
- Death by MI before age 30
- Treat w/ plasma LDL apheresis

Xanthoma
Cholesterol

1) Acetyl-CoA → HMG-CoA → Mevalonate

2) Mevalonate (C$_6$) + 3ATP → Isopentenyl-PP$_i$ (C$_5$ “isoprene”) + CO$_2$ + 3ADP + P$_i$

3) 6 Isoprene units (C$_5$) → Squalene (C$_{30}$)

4) Squalene (C$_{30}$) → Cholesterol (C$_{27}$)
Lipid Trafficking Enzymes

Lipoprotein Lipase, LPL
(located on endothelium of muscle, adipose tissue)

Lecithin:Cholesterol Acyltransferase (LCAT)
(located on HDL)

Also CETP
## Lipoprotein Particles

<table>
<thead>
<tr>
<th>Apolipoprotein</th>
<th>M.W.</th>
<th>gL$^{-1}$</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>ApoA-I</td>
<td>28 kD</td>
<td>1.0-1.2</td>
<td>CMC, HDL; activates LCAT</td>
</tr>
<tr>
<td>ApoA-II</td>
<td>17 kD</td>
<td>0.3-0.5</td>
<td>CMC, HDL</td>
</tr>
<tr>
<td>ApoA-IV</td>
<td>46 kD</td>
<td>0.15-0.16</td>
<td>CMC, HDL</td>
</tr>
<tr>
<td>ApoB-48</td>
<td>264 kD</td>
<td>0.03-0.05</td>
<td>CMC; (intestine only)</td>
</tr>
<tr>
<td>ApoB-100</td>
<td>512 kD</td>
<td>0.7-1.0</td>
<td>VLDL, IDL, LDL; binds receptor</td>
</tr>
<tr>
<td>ApoC-I</td>
<td>7 kD</td>
<td>0.04-0.06</td>
<td>CMC, VLDL, HDL</td>
</tr>
<tr>
<td>ApoC-II</td>
<td>9 kD</td>
<td>0.03-0.05</td>
<td>CMC, VLDL, HDL; activates LPL</td>
</tr>
<tr>
<td>ApoC-III</td>
<td>9 kD</td>
<td>0.12-0.14</td>
<td>CMC, VLDL, HDL</td>
</tr>
<tr>
<td>ApoD</td>
<td>33 kD</td>
<td>0.06-0.07</td>
<td>HDL</td>
</tr>
<tr>
<td>ApoE</td>
<td>38 kD</td>
<td>0.03-0.05</td>
<td>CMC, VLDL, IDL, HDL; binds receptor</td>
</tr>
</tbody>
</table>

ApoA-I Milano Clinical Trial in Humans, JAMA 290(17):2292 2003
ApoB RNAi in Mice, Nature 432:173 2004
Isopentylpyrophosphate (MEV Pathway)
The Statins

Fermentation-Derived Statins

Mevastatin

Lovastatin

Simvastatin

Pravastatin

Synthetic Statins

Atorvastatin

Fluvastatin

Rosuvastatin

Cerivastatin*

STATINS REDUCE [CIRCULATING CHOLESTEROL] BY UP TO 50%

FAIRLY SAFE (rhabdomyolysis)

SHORT HISTORY

• 1976 Mevastatin from *Penicillium citrinum*

• 1980 Mevinolin from *Aspergillus terreus*

• 1987 FDA approves Lovastatin

• 1988 Lovastatin not effective in FH homozygotes

• 1995 Pravastatin decreases heart transplant rejection and mortality independently of lowering cholesterol levels
IDEA:

Statins act by inhibiting cholesterol synthesis in patients.
Clinical Observation

Lovastatin therapy in receptor-negative homozygous familial hypercholesterolemia:
Lack of effect on low-density lipoprotein concentrations or turnover

Ricardo Uauy, MD, PhD, Gloria Lena Vega, PhD, Scott M. Grundy, MD, PhD,
and David M. Bilheimer, MD

From the Departments of Pediatrics, Internal Medicine, and Clinical Nutrition, and Center for
Human Nutrition, University of Texas Southwestern Medical Center at Dallas

To determine whether at least part of the fall in low density lipoprotein (LDL)
levels during lovastatin therapy might be the result of a reduced secretion of
lipoproteins by the liver, three children 6 to 9 years of age with receptor-
negative homozygous familial hypercholesterolemia underwent treatment
with lovastatin. These patients have no capacity to synthesize LDL receptors.
During lovastatin therapy, at a dose of 2 mg/kg/day, there was no decrease in
LDL-cholesterol levels, nor was the turnover rate of LDL affected by the drug. The
only significant change was a 74% drop in very low-density lipoprotein during
treatment. We conclude that lovastatin is not effective in treatment of receptor-
negative homozygous familial hypercholesterolemia. The most likely mecha-
nism of action for this drug is to increase LDL receptor activity. (J Pediatr
1988;113:387-92)

Sterol Regulation of Transcription by SREBP

SREBP Transcription Factor Localization

SCAP/Insig proteins also (HMG CoA R)

ER Retention

Proteases
IDEA:

Statins act by activating SREBP, resulting in increased LDL receptor and more efficient recruitment of LDL out of plasma.
Clinical Observations:  
The Pleiotrophic Effects of Statins

- Statins reduce organ rejection and mortality after cardiac transplant. Randomized, placebo-controlled
  

- Statins reduce inflammation in patients with chronic rheumatoid arthritis. Randomized, placebo-controlled
  

- Statins reduce incidence of MS lesions. Small open-label clinical trial.
  

- Statins reduce proteinuria in systemic lupus erythematosus. Small open-label clinical trial.
  
Multiple Sclerosis

- Effects up to one million worldwide
- 16,000 deaths in 2002 (0.03 %)
- Autoimmune attack of myelin in brain and spinal cord
- Progressive physical disability
- Current therapies include injected Ifn-β and copaxone (basic peptides)

Lesions on cerebellum and spinal cord of an MS patient; Jean Cruvelhier circa 1860
Immune System Overview

Inate

NK cells

Adaptive

APC

B cells

T cells

γδ

professional

CD4 helper

CD8 cytotoxic

T_1

T_2

Ifn-γ

IL2

Ifn-γ

TNFα

IL4, 10

TGFβ

Ifn-β

Ifn-γ

induced

IL12 from activated macrophages and dendritic cells

+
Cell Mediated Adaptive Immune Response
Tc/MHC1/CD8 versus Th/MHC2/CD4
MHC Antigen Presentation

Expressed in ALL CELLS

- Class I MHC
- Exogenous antigen
- Endosomal pathway
- Class II MHC

Expression of MHC antigens can be induced by INF-γ during acute immune response; this is also a hallmark of auto-immune diseases.

INF-γ can induce expression in non-APC cells during acute immune response; this is also a hallmark of auto-immune diseases.
The MHC Molecules

Class I

Class II
Different Responses to Different Pathogens

<table>
<thead>
<tr>
<th>Typical pathogens</th>
<th>Cell-mediated immunity</th>
<th>Humoral immunity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vaccinia virus</td>
<td>Mycobacterium tuberculosis</td>
<td></td>
</tr>
<tr>
<td>Influenza virus</td>
<td>Mycobacterium leprae</td>
<td></td>
</tr>
<tr>
<td>Rabies virus</td>
<td>Leishmania donovani</td>
<td></td>
</tr>
<tr>
<td>Listeria</td>
<td>Pneumocystis carinii</td>
<td></td>
</tr>
<tr>
<td>Clostridium tetani</td>
<td>Staphylococcus aureus</td>
<td></td>
</tr>
<tr>
<td>Streptococcus pneumoniae</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Polio virus</td>
<td>Pneumocystis carinii</td>
<td></td>
</tr>
<tr>
<td>Trichinella spiralis</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Location</th>
<th>Cytosol</th>
<th>Macrophage vesicles</th>
<th>Extracellular fluid</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Effector T cell</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Cytotoxic CD8 T cell</td>
<td></td>
<td>T&lt;sub&gt;H&lt;/sub&gt;1 cell</td>
<td>T&lt;sub&gt;H&lt;/sub&gt;1 and T&lt;sub&gt;H&lt;/sub&gt;2 cells</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Antigen recognition</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptide:MHC class I complex on infected cell</td>
<td>Peptide:MHC class II complex on infected macrophage</td>
<td>Peptide:MHC class II complex on antigen-specific B cell</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Effector action</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Killing of infected cell</td>
<td></td>
<td>Activation of infected macrophages</td>
<td>Activation of specific B cell to make antibody</td>
</tr>
</tbody>
</table>
Th1 vs. Th2 Helper Cells

- Site of infection: extracellular pathogens, e.g., helminths, also oral antigens

Th1 cells are activated by microbial infection, leading to the production of IL-12, which activates Th1 cells. Effector Th1 cells contribute to the elimination of extracellular pathogens.

Th2 cells are activated by antigen-presenting cells, leading to the production of cytokines such as IL-4, which activate B cells to produce antibodies. Effector Th2 cells contribute to the elimination of extracellular pathogens.

LFA-1 is a key molecule in the interaction between Th1 and Th2 cells, facilitating cell-cell contact and immune response.
Th1 Helper Cells (macrophage/inflammatory)

**IN PERIPHERAL LYMPHOID ORGAN**

- Mature dendritic cell with ingested bacteria
- B7 ligand
- IL-12 receptor
- CD28

**NAIVE HELPER T CELL**

- Naive helper T cell

**INFECTED DENDRITIC CELL STIMULATES NAIVE HELPER T CELL TO DIFFERENTIATE INTO EFFECTOR TH1 CELL**

**AT SITE OF INFECTION**

- Effector TH1 cell
- CD40 ligand
- CD40

**INTERFERON-γ RECEPTOR**

- Infected macrophage
- Killed bacteria

**EFFECTOR TH1 CELL ACTIVATES INFECTED MACROPHAGE TO KILL THE INTRACELLULAR BACTERIA**

**migration**
Th2 Helper Cells (B cell response)

Also mast cell degranulation and eosinophil activation high [antigen], nonprofessional
Th1 vs. Th2 Antagonism

STATINS ALTER THIS BALANCE IN FAVOR OF TH2 HELPER CELLS. BUT HOW?
Statins Bind to LFA-1 Directly
Isoprenoids in Humans

1. Steroids Hormones
2. Metabolites (Vit. A, E, K; co-Q; 25,000 terpenoids)

3. Isopentyl adenosine (tRNA)
4. Dolichol (N-linked glycosylation)
5. Protein Prenylation
Isoprenoids in G Protein Signaling

Heterotrimeric (GPCR’s)

“Small” (RTK’s)
How Do Statins Work?

Reported to:

- Decrease occurrence of AD by 70% in retrospective study
- Suppress MS (clinical trials)
- Block SMC proliferation/migration in vitro
- Modulate NF-κB Function in vitro
- Block IFN-γ induced MHCI expression in vitro
- Suppress expression/secretion of immunoinflammatory molecules in vitro
- Direct inhibition of LFA-1

Most in vitro effects reversed by mevalonate, transF, transGG, but not cisF, cisGG, squalene, isopentyl adenosine or ubiquinone; prenylation inhibitors have many of the same immunomodulatory effects that are observed for statins.

