M1 - Immunology, Winter 2008

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Arachidonic Acid Metabolites and Inflammation

Joseph Fantone, M.D.
Host Defense 2/12  10-11:00am
INFLAMMATORY MEDIATORS

PLASMA DERIVED
• COMPLEMENT CASCADE
  C3a, C5a
• COAGULATION CASCADE
  Thrombin, plasmin

CELL-DERIVED
• VASOACTIVE AMINES
  histamine, serotonin
• OXYGEN METABOLITES
  hydrogen peroxide (H₂O₂)
  superoxide anion (O₂⁻)
  hypochlorous acid (HOCl⁻)
• ARACHIDONIC ACID METABOLITES
  cyclooxygenase-derived
  lipoxygenase-derived
• CYTOKINES
  Interleukins
  Chemokines
  Interferons
  Tumor Necrosis Factor
  Growth Factors
Intended Learning Outcomes: To Understand The

- Primary inflammatory mediators derived from the metabolism of arachidonic acid including their primary cellular source and biological activity.

- Effects of nonsteroidal anti-inflammatory compounds on blocking the production of arachidonic acid metabolites during disease.

- Mechanism of aspirin therapy and diets rich in fish containing high levels of omega 3 fatty acids as potentially important in lowering the incidence of cardiovascular disease.
YOU ARE WHAT YOU EAT
Arachidonic acid

Phospholipid

Phospholipase A

Lysophospholipid

Phospholipase C

Arachidonic acid + phospholipase C

Diacylglycerol

Diacylglyceride lipase

Cyclooxygenase 1 + Lipoxygenase Products

Cyclooxygenase 2
Cell Membrane Phospholipids

\[ \text{Arachidonic Acid} \]

\[ \text{PGG}_2 = \text{PGH}_2 \]

\[ \text{TXA}_2 = \text{UNSTABLE} \]

\[ \text{PGF}_{2\alpha} \]

\[ \text{6-Keto PGF}_{1\alpha} \]

LIPID PATHWAY

HETEs [mono & di]

LEUKOTRIENE [SRS-A]
Leukotriene Synthesis

Arachidonic Acid $\rightarrow$ 5-HPETE $\rightarrow$ Leukotriene A (LTA)

Lipoxygenase

Glutathione-S-transferase

Leukotriene B (LTB) $\rightarrow$ Leukotriene C (LTC) $\rightarrow$ Leukotriene D (LTD)
<table>
<thead>
<tr>
<th>CELL</th>
<th>PRODUCT</th>
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</thead>
<tbody>
<tr>
<td>Neutrophils</td>
<td>Leukotrienes</td>
</tr>
<tr>
<td>Macrophage/Monocyte</td>
<td>Prostaglandins + Leukotrienes</td>
</tr>
<tr>
<td>Platelets</td>
<td>Thromboxane</td>
</tr>
<tr>
<td>Endothelial Cells</td>
<td>Prostacyclin</td>
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</tbody>
</table>
## Biological Function

### Cyclooxygenase-derived Products:

<table>
<thead>
<tr>
<th>Prostaglandin E₂/Prostacyclin</th>
<th>Immunoregulatory</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>• Inhibits Immune cell activation</td>
</tr>
<tr>
<td></td>
<td>• Inhibits cytokine production</td>
</tr>
<tr>
<td></td>
<td>• Inhibits mast cell activation</td>
</tr>
<tr>
<td></td>
<td>Blocks platelet aggregation</td>
</tr>
<tr>
<td></td>
<td>Increases vasodilation</td>
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<td>Stimulates adenylate cyclase</td>
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</table>

<table>
<thead>
<tr>
<th>Thromboxane</th>
<th>Causes vasoconstriction</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Induces platelet aggregation</td>
</tr>
</tbody>
</table>
Biological Function

Lipoxygenase-derived Products:

Leukotriene $B_4$  Neutrophil Activation
- degranulation

Mast cell activation
- degranulation

Leukotriene C,D,E (SRS-A)  Causes smooth muscle contraction
Increases vascular permeability
In Vivo Effects of Arachidonic Acid Derived Products

- Regulates Thermostatic Set Point (Fever)
- Regulates Pain (Interacts with pain receptors)
- Regulates Blood Flow
- Regulates Leukocyte Activity
Hypothalamus

Viruses
Bacteria
Toxins

Activated leukocytes
Endogenous pyrogen

Phagocytic leukocytes

Arachidonic Acid
Prostaglandin E2
Temperature

Aspirin
NSAIDs

(e.g. Interleukin-1)

Shivering
Sweating
Vasomotor tone
Rheumatoid Arthritis distorts joints

Source: http://www.nih.gov/
Immunopathology of Rheumatoid Arthritis

- Complement
  - Anti-altered IgG
  - Fixation Activation
  - Chemotaxis
  - Lysosomal Enzymes
    - Collagenase
    - Neutral Proteases
    - Phospholipase

- Activated oxygen
  - $O_2$, $H_2O_2$

- Nonsteroidal Anti-inflammatory Agents
- Arachidonic Acid
  - Prostaglandins
  - Nerve Sensitization
  - Vasodilation

Cartilage
- Subchondral bone plate

Source: Undetermined
Chemotactic Activity of LTB4

BY: Greg Luerman

GNU 1.2

Pharmacologic Regulation of Arachidonic Acid-Derived Products

• Modulate Phospholipase activity:
  – Suppress the release of arachidonic acid (no substrate available)
  – Blocks both COX and LO-derived products

• Modulate Cyclooxygenase Activity:
  – Blocks Cyclooxygenase-derived products
  – COX-1 and COX-2 inhibitors

• Modulate specific enzymes down-stream from COX:
  – Thromboxane synthetase inhibitors

• Modulate lipoxygenase activity:
  – Block 5-lipoxygenase enzyme
  – Small molecule receptor antagonists for cysteinyi leukotrienes
Non-Steroidal Anti-Inflammatory Compounds

- Aspirin (acetylsalicylic acid)
- Ibuprofen (propionic acid derivatives)
- Indomethacin (indole derivatives)
- Tylenol (Acetaminophen)
- COX-2 Inhibitors (Vioxx, celebrex, Bextra)
COX-2 Inhibitors

- **CELEBREX** (Celecoxib) Pfizer-(Pharmacia)
- **BEXTRA** (Valdecoxib) Pfizer
- **VIOXX** (Rofecoxib) Merck

Osteoarthritis
Rheumatoid arthritis
Primary dysmenorrhea
Pain management
Complications!!
INHIBITS CYCLO-OXYGENASE ENZYME IRREVERSIBLY BY ACETYLATED THE ENZYME AT THE ACTIVE SITE, THUS THE PRODUCTION OF ENDOPEROXIDES AND THEIR DERIVATIVES, INCLUDING PROSTAGLANDINS, THROMBOXANES, AND PROSTACYCLINS WILL BE INHIBITED.
BOTH INHIBIT CYCLO-OXYGENASE ACTIVITY BY BINDING REVERSIBLY TO THE ACTIVE SITE OF THE ENZYME, THUS BLOCKING THE FORMATION OF PROSTAGLANDINS, THROMBOXANES, AND PROSTACYCLINS.
AN ASPIRIN A DAY

Roughly 80 million aspirin tablets are consumed daily in the USA
Of those:
72% are taken for disease prevention
28% are taken for pain
Reduce the risk of heart attack or stroke with……

Aspirin

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THE HOMEOSTATIC BALANCE

PGI₂
ENDOTHELIUM

TXA₂
PLATELETS
Thrombus Formation

- Elastic Lamina
- Platelets
- Basement Membrane
- Thromboxane
- ADP
- Collagen
- Injury
- Aggregation
- Organization
- Plaque
Can Aspirin Act As An Anti-thrombogenic Agent?

- Inhibits platelet aggregation by blocking platelet-derived thromboxane production

- Blocks platelet cyclooxygenase for the life of the platelet, as no new protein synthesis occurs

- Blocks endothelial cell-derived prostacyclin

- Suppression of endothelial cell-derived prostacyclin is short lived as endothelial cells can generate new cyclooxygenase enzyme

- Platelet activity is blocked more than endothelial cell activity
COX-2 inhibitors work by blocking COX-2 enzyme which is involved in the inflammation pathway. By sparing COX-1 gastrointestinal toxicity is reduced.

Physiologic Stimuli → COX-1 (constitutive) → Prostaglandin E_2 (Renal function) → Thromboxane A_2 (Platelet function) → Prostacycline (PGI_2) (Gastric Protection)

COX-2 (inducible) → Pro-inflammatory PGs and other inflammatory mediators → Inflammation

Inflammatory Stimuli
lipid mediators of Inflammation

Stimulus

Cell membrane
Phospholipids

+ Phospholipase

Arachidonic acid
Acute inflammation: lipid mediators

Stimulus

Phospholipase

Cell membrane
Phospholipids

Arachidonic acid

COX-1+2
Prostaglandins
Prostaglandin E₂
Prostacyclin PGI₂

COX-1
Thromboxanes
TXB₂

Lipooxigenases (5-LO)
Leukotrienes
LTB₄
LTC₄, LTD₄
Arachidonic acid

Cell membrane
Phospholipids

Stimulus

+ Phospholipase

Arachidonic acid

COX-1+2
Prostaglandins
Prostaglandin E$_2$
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COX-1
Thromboxanes
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Lipooxigenases (5-LO)
Leukotrienes
LTB$_4$
LTC$_4$, LTD$_4$

Vasodilation  Increase vascular permeability  Control platelet aggregation  Chemotaxis  Pain  Fever
Acute inflammation: lipid mediators

An important role in vascular homeostasis

Endothelium

Prostacyclin PGI$_2$  $\rightarrow$  TXB2

Platelets

Anti-thrombotic  $\leftrightarrow$  Pro-thrombotic
Prostacyclin PGI$_2$ is an anti-thrombotic mediator produced by the endothelium. TXB2 is a pro-thrombotic mediator produced by platelets. NSAIDs inhibit both COX-1 and COX-2; COXIBs inhibit COX-2.
Acute inflammation: lipid mediators

- **Therapeutic targets**
  - **Endothelium**
    - COX-2
    - Prostacyclin PGI$_2$
    - Anti-thrombotic
  - **Platelets**
    - COX-1
    - TXB2
    - Pro-thrombotic

* Classical NSAID, it inhibits both COX enzymes
Acute inflammation: lipid mediators

Therapeutic targets

Endothelium

Platelets

COX-2

Prostacyclin PGI₂

TXB₂

COX-1

Anti-thrombotic

Pro-thrombotic

Vioxx®
Acute inflammation: lipid mediators

Therapeutic targets

Aspirin inhibits COX-2 irreversibly

Endothelium

Aspirin inhibits COX-1 irreversibly

Platelets

Prostacyclin PGI₂

All cells but the platelet can resynthesize the enzymes

TXB₂

Anti-thrombotic

Pro-thrombotic
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